Supplementary Information

Encoding BRAF Inhibitor Functions in Protein Degraders

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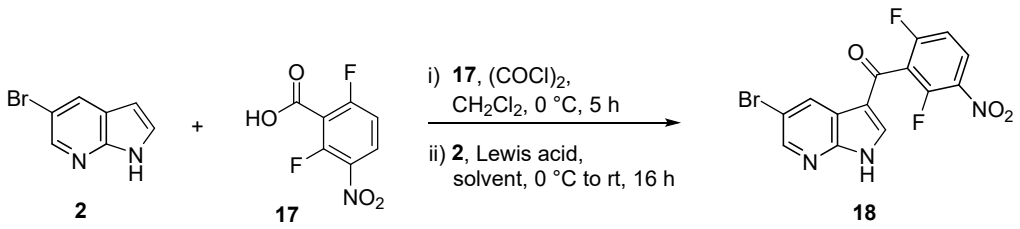


2	**17**	Lewis acid	solvent	yield **18** [a]
1.0 Eq.	1.0 Eq.	5.0 Eq. $AlCl_3$ [b]	dry CH_2Cl_2	13%
1.0 Eq.	1.0 Eq.	**5.0 Eq. $ZrCl_4$** [c]	dry CH_2Cl_2	10%
1.0 Eq.	1.0 Eq.	5.0 Eq. $AlCl_3$	**1,2-DCE**	9%
1.0 Eq.	**4.0 Eq.**	5.0 Eq. $AlCl_3$	dry CH_2Cl_2	4%
1.5 Eq.	1.0 Eq.	5.0 Eq. $AlCl_3$	dry CH_2Cl_2	31%
1.5 Eq.	1.0 Eq.	**7.5 Eq. $AlCl_3$**	dry CH_2Cl_2	49%

Table S1: Reaction optimization for the key entry step towards functionalized azaindoles. [a] Isolated yields after purification by column chromatography. [b] It has been suggested that azaindoles are highly coordinated by at least three molecules $AlCl_3$.[1] Accordingly, a high excess of Lewis acid is needed to accelerate the reaction. [c] Guchhait *et al.*[2] reported on the $ZrCl_4$-mediated regio- and chemoselective Friedel-Crafts acylation of indoles.

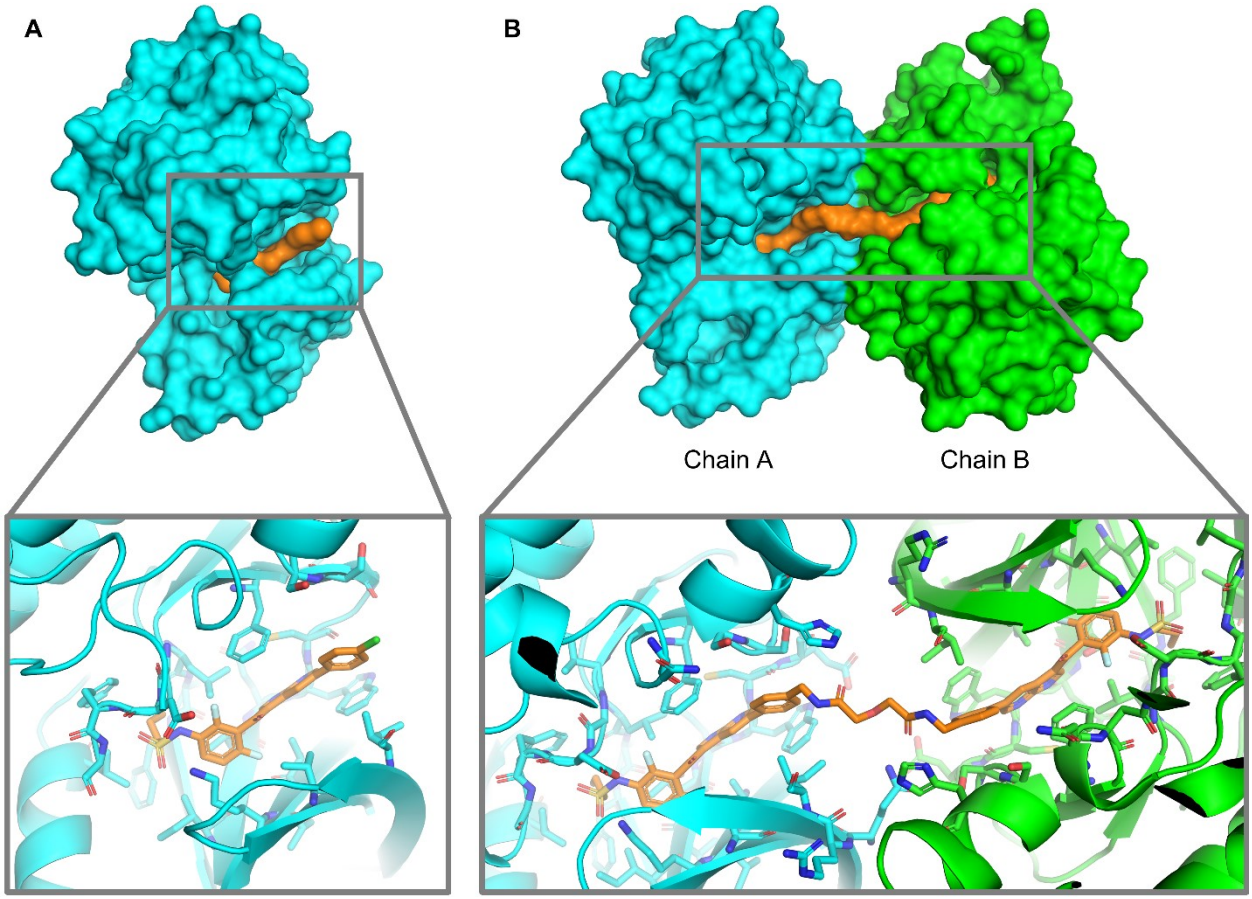


Figure S1: $BRAF^{V600E}$ in complex (A) with vemurafenib (PDB 3OG7)[3] and (B) with a homodimeric vemurafenib derivative (PDB 5JT2).[4]

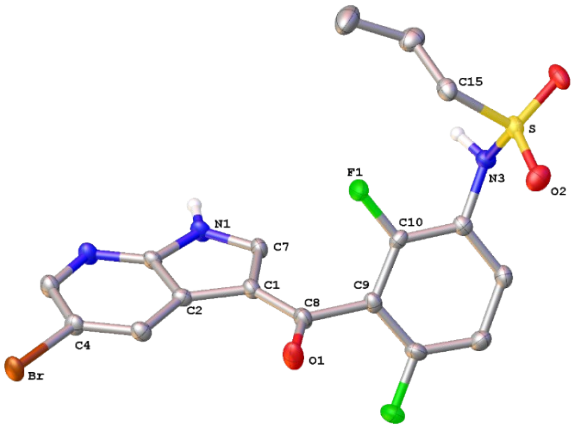


Figure S2: Molecular plot of the X-ray crystal structure of acylated azaindole **3** demonstrating that the Friedel-Crafts acylation occurred at the correct position.

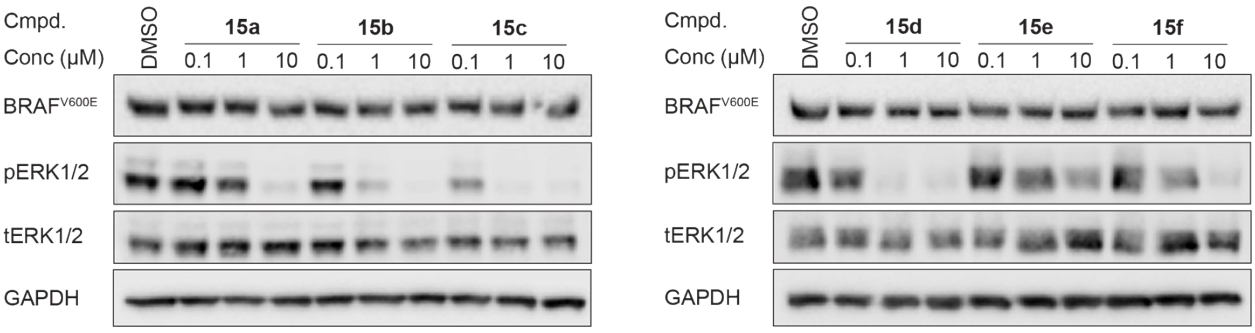


Figure S3: PROTACs **15a** to **15f** inhibit ERK signalling in SK-MEL-28 cells, but do not degrade $BRAF^{V600E}$. Cells were treated with compounds for 4 h at the concentration indicated.

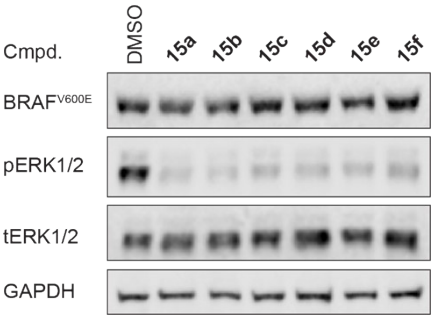


Figure S4: Experiment with prolonged treatment time. SK-MEL-28 cells were treated with 10 μM of the compounds for 24 h.

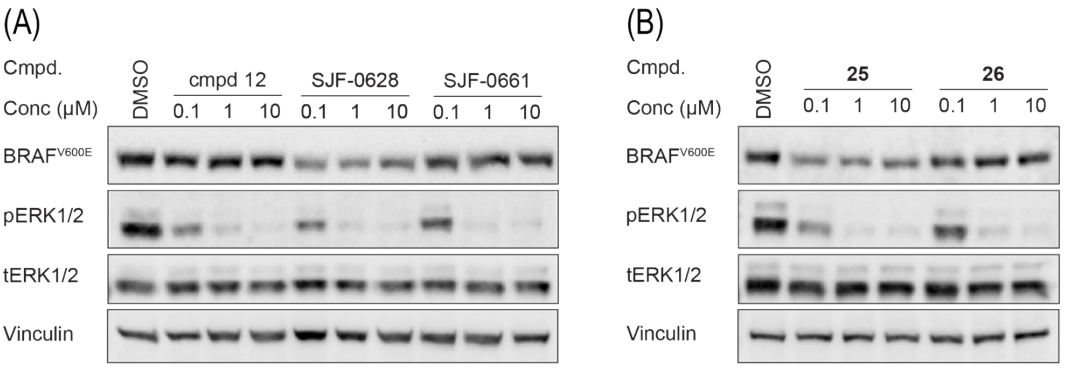


Figure S5: Vemurafenib-based PROTACs were evaluated against paradox breaker-based PROTACs in SK-MEL-28 cells. (A) The CRBN-hijacking PROTAC cmpd 12, the VHL-targeting PROTAC SJF-0626, as well as its negative control (SJF-0661) were used to treat SK-MEL-28 cells for 4 h at the dose indicated; (B) The paradox-breaker PROTAC **25** (PLX7683-based), but not compound **26** (PLX7904-based) cause $BRAF^{V600E}$ degradation in SK-MEL-28 cells after a 4 h treatment at the concentration indicated.

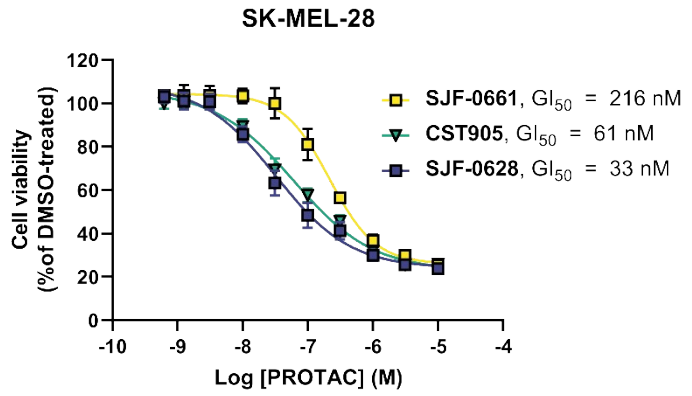


Figure S6: CellTiter-Glo luminescent cell viability assay performed in SK-MEL-28 cells after 120 h treatment with the indicated compounds. Data is represented as mean±s.d. (n=3).

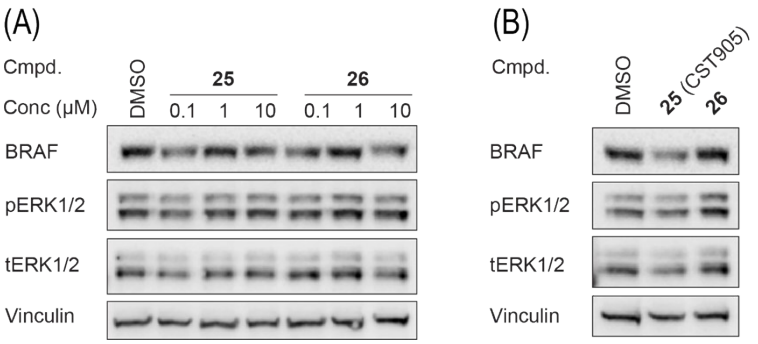


Figure S7: Paradox-breakers **25** and **26** do not enhance ERK signaling in the NRAS mutant cell line SK-N-AS. (A) Cells were treated with PROTACs for 4 h at the dose indicated. (B) Prolonged treatment time (24 h) and increased concentration (10 μM) was used.

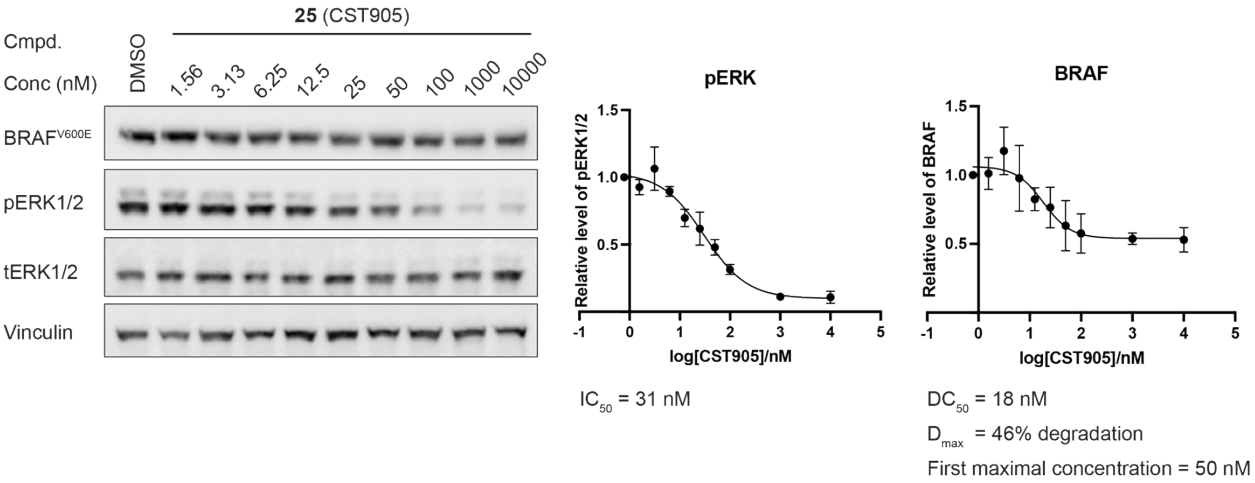
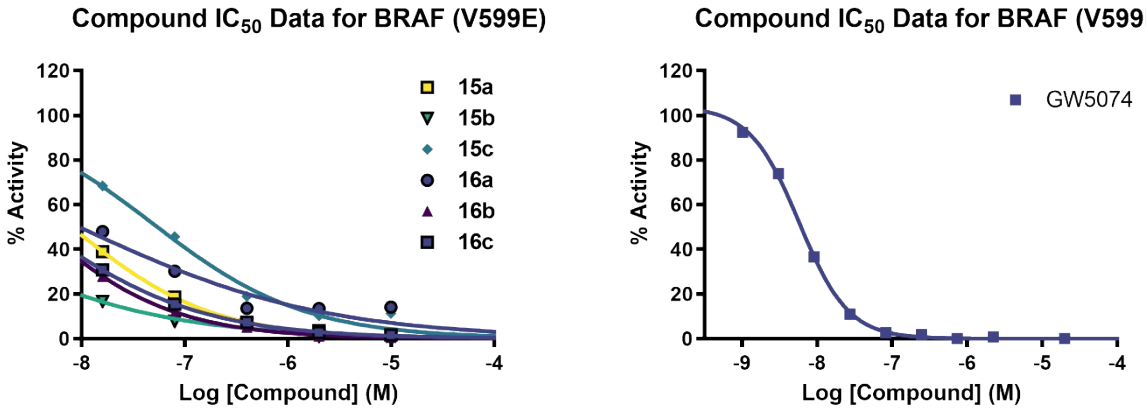


Figure S8: Dose-dependent analysis of BRAF^{V600E} degradation and inhibition of ERK phosphorylation in SK-MEL-28 cells. Cells were treated for 4 h with PROTAC **25** (CST905) at the dose indicated. Quantified values represent mean of five independent repeats.



	15a	15b	15c	16a	16b	16c	GW5074
Hill Slope	–0.6493	–0.4317	–0.5875	–0.3718	–0.6447	–0.5497	–1.321
IC_{50} (nM)	8.5	0.31	48	10	3.4	3.8	5.8

Figure S9: Determination of compound inhibition of BRAF V599E (HGNC BRAF V600E) in a radiometric functional kinase assay (https://www.reactionbiology.com/services/kinase-assays/kinase-screening). Single determinations of IC_{50} with 10 µM [^{33}P]-ATP and 1 µM substrate peptide. Curve fits to estimate IC_{50} were performed where the enzyme activities at the highest concentration of compounds were less than 65%.

Supplementary Information: Biochemistry

A. Kinase inhibition assay

Compounds were assayed for inhibition of BRAF V599E (HGNC reference BRAF V600E) kinase activity in a commercial screening service (HotSpot™, Reaction Biology Corporation, Malvern, PA, USA) https://www.reactionbiology.com/services/kinase-assays/kinase-screening (accessed 13 Feb 2022). In brief, substrate peptide (MEK1 (K97R), 1 μM) phosphorylation was quantified following transfer of [^{33}P]-labelled phosphate from radio-labelled ATP (10 μM) using a filter membrane protocol.[5] Compounds were tested in 5-dose IC_{50} mode with a 5-fold serial dilution starting at 10 μM (see Figure S9). Control compound, GW5074, was tested in 10-dose IC_{50} mode with 3-fold serial dilution starting at 20 μM. Curve fits were performed where the enzyme activities at the highest concentration of compounds were less than 65%.

B. Cell lines

The human melanoma cell line SK-MEL-28 and the human neuroblastoma cell line SK-N-AS were used throughout this study. SK-MEL-28 were a gift from Cell Services at the Francis Crick Institute. SK-N-AS cells were obtained from ECACC. Both cell lines were certified negative for mycoplasma and were authenticated by short tandem repeat profiling. Both cell lines were maintained in Dulbecco's Modified Eagle Medium (Thermo Fisher Scientific), supplemented with 10% fetal calf serum (FCS, Thermo Fisher Scientific), and incubated at 37°C and 5% CO_2.

C. Immunoblotting

For immunoblotting, cells were plated at a density of 100,000 cells per well in 6 well plates (Falcon), incubated for 48h and treated as indicated. Cells were lysed in D0.4 lysis buffer (20 mM HEPES pH 7.5, 10% Glycerol, 0.4 M KCl, 0.4% Triton X-100, 15 mM EDTA) supplemented with cOmplete Mini Protease Inhibitor Cocktail (Sigma Aldrich) and PhosSTOP phosphatase inhibitors (Sigma Aldrich) and clarified lysates run on NuPAGE 4-12% Bis-Tris gels before transfer to Immobilon-P or -FL PVDF membrane (Merck Millipore). Immunoblotting was performed with the following antibodies: B-RAF (Cell Signaling Technology, Cat. # 14814, 1:1000), Phospho-ERK1/2 (Cell Signaling Technology, Cat. # 4370), ERK1/2 (Cell Signaling Technology, Cat # 4696, 1:1000), Phospho-MEK1/2 (Cell Signaling Technology, Cat. # 9154, 1:1000), MEK (Cell Signaling Technology, Cat. # 9122, 1:1000), GAPDH (Cell Signaling Technology, Cat. #

97166, 1:5000) and Vinculin (Sigma V9624, 1:5000). Western blots were visualised using a LI-COR Odyssey and quantified with ImageJ. For quantifications, densitometry measurements were normalised to loading controls and are shown relative to DMSO-treated cells. DC_{50} calculation was performed using GraphPad Prism, fitting a curve to the data using the log(inhibitor) vs. response -- variable slope (four parameters) function.

D. Cell viability assay

For cell viability assays, SK-MEL-28 cells were plated at 400 cells per well in wells of two white 384-well plates (Corning). After 24h, a CellTiter-Glo® Luminescent Cell Viability assay was performed on one plate to give T_0 readings. On the other plate, an Echo 650 Liquid Handler (Labcyte) was used to dispense compounds at the indicated final concentrations. Cells were incubated for 5 days and a CellTiter-Glo® Luminescent Cell Viability assay was performed. Luminescence readings were normalised to DMSO controls and curve fitting and GI_{50} calculation performed with GraphPad Prism, using the log(inhibitor) vs. response -- Variable slope (four parameters) function.

E. Computational studies

Figures 2 and S1 were created using PyMOL (The PyMOL Molecular Graphics System, Version 2.5.2, Schrödinger, LLC).

F. Molecular descriptor calculations

Predicted values for the topological polar surface area (TPSA) were calculated using SwissADME (Swiss Institute of Bioinformatics).[6]

G. logD measurements

The determination of the $logD_{7.4}$ values was performed by a chromatographic method as described previously.[7–9] The system was calibrated by plotting the retention times of six different drugs (atenolol, metoprolol, labetalol, diltiazem, triphenylene, permethrin) versus their literature known $logD_{7.4}$ in a calibration line ($R^2 = 0.99$). Subsequently, the mean retention times of the analytes were taken to calculate their $logD_{7.4}$ values with aid of the calibration line. At least two independent measurements of each analyte were performed.

H. Plasma protein binding studies

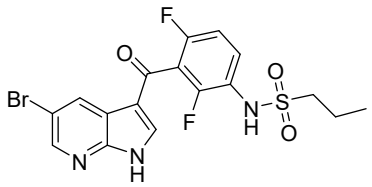
Plasma protein binding (%PPB) was estimated by correlating the logarithmic retention times of the analytes on a CHIRALPAK HSA 50 × 3 mm, 5 µm column with the literature known %PPB values (converted into logK values) of the following drugs: warfarin, ketoprofen, budesonide, nizatidine, indomethacin, acetylsalicylic acid, carbamazepine, piroxicam, nicardipine, and cimetidine (for details, see Valko *et al.*[10]). Samples were dissolved in MeCN/DMSO 9:1 to achieve a final concentration of 0.5 mg/mL. The mobile phase A was 50 mM ammonium acetate adjusted to *p*H 7.4 with ammonia solution, while mobile phase B was *i*PrOH. The flow rate was set to 1.0 mL/min, the UV detector was set to 254 nm, and the column temperature was kept at 30 °C. After injecting 2 µL of the sample, a linear gradient from 100% A to 30% *i*PrOH in 5.4 min was applied. From 5.4 to 18 min, 30% *i*PrOH was kept, followed by switching back to 100% A in 1.0 min and a re-equilibration time of 6 min. With the aid of the calibration line ($R^2 = 0.94$), the logK values of new substances were calculated and converted to their %PPB values. At least two independent measurements of each analyte were performed.

I. Synthesis: general remarks

Preparative column chromatography was performed using Merck silica gel 60 (0.063 – 0.200 mm) or using an automated flash chromatography system puriFlash XS 520Plus. Melting points were determined on a Büchi 510 oil bath apparatus or on a Reichelt hot-stage apparatus and were uncorrected. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker Avance 500 MHz NMR spectrometer or on a Bruker Avance III 600 MHz NMR spectrometer, respectively. NMR spectra were processed and analyzed in MestReNova. Chemical shifts are given in parts per million (ppm), coupling constants *J* are given in Hertz, and spin multiplicities are given as s (singlet), d (doublet), t (triplet), q (quartet) or m (multiplet). In case of overlapping extraneous solvent peaks, multiplet analyses in ^{1}H NMR spectra were performed using qGSD (quantitative Global Spectral Deconvolution). Resonance assignments were made on the basis of one- and two-dimensional NMR techniques which include ^{1}H, ^{13}C, DEPT, HSQC, and HMBC experiments. HRMS was recorded on a micrOTOF-Q mass spectrometer (Bruker) with ESI-source coupled with an HPLC Dionex UltiMate 3000 (Thermo Scientific). The purity and identity of compounds were determined on an Infinity Lab LC/MSD-system (Agilent) with ESI-source coupled with a HPLC 1260 Infinity II (Agilent) using a EC50/2 Nucleodur C18 Gravity 3 µm column (Macherey-Nagel). The column temperature was 40 °C. HPLC conditions started with 90% H_2O containing 2 mM NH_4Ac. The gradient ramped up to 100% MeCN in 10 min, followed by further flushing with 100% MeCN for 5 min. The flow rate was 0.5 mL/min. The samples were dissolved in H_2O, MeOH or MeCN (approx. 1 mg/mL), and 2 µL sample solution was injected. Positive total ion scans were observed from 100–1000 *m/z* (or more if necessary) and UV absorption was detected from 190–600 nm using a diode array detector (DAD). The purity was determined at 220–600 nm.

J. Synthesis: procedures

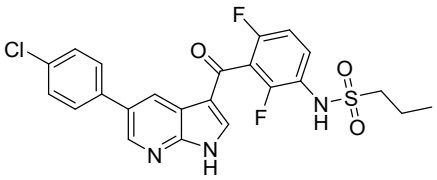
***N*-[3-(5-Bromo-1*H*-pyrrolo[2,3-*b*]pyridine-3-carbonyl)-2,4-difluoro-phenyl]propane-1-sulfonamide (3)**[11]



2,6-Difluoro-3-(propylsulfonamido)benzoic acid (**1**, 5.58 g, 20 mmol) was dissolved in dry CH_2Cl_2 (100 mL). Oxalyl chloride (2M in CH_2Cl_2, 30 mL, 60 mmol) and dry DMF (5 drops) were added and the mixture was stirred under argon atmosphere for 5 h. The solvent was removed under reduced pressure. 5-Bromo-7-azaindole (**2**, 5.92 g, 30 mmol) was suspended in dry CH_2Cl_2 (150 mL) and $AlCl_3$ (20.0 g, 150 mmol) was added in portions at 0 °C. Subsequently, the *in situ* generated acid chloride in dry CH_2Cl_2 (50 mL) was added dropwise, and the reaction mixture was stirred for 18 h under argon and was allowed to warm up to rt slowly. Subsequently, the mixture was poured onto a mixture of ice-cold brine and saturated $NaHCO_3$ solution. It was extracted with EtOAc (3 × 300 mL), washed with H_2O (200 mL) and brine (200 mL), dried over Na_2SO_4, filtered and evaporated. After purification by column chromatography (gradient of petroleum ether/EtOAc 1:1 to EtOAc), **3** was obtained as a pale brown solid.

Yield (6.42 g, 70%); R_f = 0.49 (petroleum ether/EtOAc 1:1); mp 242 – 244 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.96 (t, *J* = 7.4 Hz, 3H), 1.75 (hept, *J* = 8.3, 7.5 Hz, 2H), 3.17 – 3.10 (m, 2H), 7.28 (t, *J* = 8.7 Hz, 1H), 7.59 (td, *J* = 9.0, 5.7 Hz, 1H), 8.27 (s, 1H), 8.50 (d, *J* = 2.3 Hz, 1H), 8.58 (s, 1H), 9.75 (s, 1H), 13.10 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 16.79, 53.47, 112.06 – 112.67 (m), 114.25, 114.93, 117.45 – 118.21 (m), 119.01, 121.84 – 122.09 (m), 128.92 (d, *J* = 9.0 Hz), 131.09, 139.30, 145.28, 147.78, 152.28 (dd, *J* = 8.2, 249.8 Hz), 155.96 (dd, *J* = 6.7, 246.8 Hz), 180.62; **^{19}F NMR** (565 MHz, DMSO-d_6) δ –117.07, –122.33; **LC-MS** (ESI) t_R = 11.02 min, 97% purity, *m/z* $[M + H]^+$ calcd for $C_{17}H_{15}{}^{81}BrF_2N_3O_3S$, 460.00; found, 459.9.

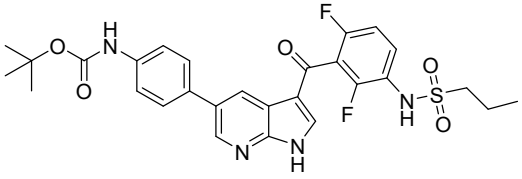
***N*-[3-[5-(4-Chlorophenyl)-1*H*-pyrrolo[2,3-*b*]pyridine-3-carbonyl]-2,4-difluorophenyl]propane-1-sulfonamide (4, vemurafenib)**[3]



Compound **3** (458 mg, 1 mmol) and 4-(chlorophenyl)boronic acid (236 mg, 1.5 mmol) were suspended in MeCN (3 mL). After the addition of Na_2CO_3 solution (2M, 2 mL), the mixture was flushed with argon and Pd(dppf)Cl_2 × CH_2Cl_2 (82 mg, 0.1 mmol) was added. It was heated *via* microwave irradiation at 120 °C for 5 min. Subsequently, the mixture was poured onto H_2O (100 mL) and it was extracted with EtOAc (3 × 50 mL). The organic layers were washed with brine (50 mL), dried over Na_2SO_4, filtered and evaporated. The crude product was purified by flash chromatography (gradient of 0 to 7% MeOH in CH_2Cl_2) to give vemurafenib (**4**) as a colourless solid.

Yield (197 mg, 40%); R_f = 0.35 (petroleum ether/EtOAc 1:1); mp 256 – 258 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.97 (t, *J* = 7.4 Hz, 3H), 1.70 – 1.79 (m, 2H), 3.10 – 3.16 (m, 2H), 7.28 (td, *J* = 1.4, 8.8 Hz, 1H), 7.54 – 7.59 (m, 2H), 7.56 – 7.63 (m, 1H), 7.79 (d, *J* = 8.5 Hz, 2H), 8.24 (s, 1H), 8.64 (s, 1H), 8.71 (d, *J* = 2.2 Hz, 1H), 9.75 (s, 1H), 13.01 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 16.80, 53.49, 112.32 (d, *J* = 22.1 Hz), 115.69, 117.47, 117.90 – 118.42 (m), 121.93 (d, *J* = 10.8 Hz), 127.06, 128.78 (d, *J* = 9.7 Hz), 128.91, 129.07, 130.25, 132.51, 137.01, 138.87, 143.93, 148.99, 152.33 (dd, *J* = 8.7, 249.7 Hz), 156.01 (dd, *J* = 6.7, 246.3 Hz), 180.63; **^{19}F NMR** (565 MHz, DMSO-d_6) δ –117.13, –122.38; **LC-MS** (ESI) t_R = 11.68 min, 97% purity, *m/z* [M + H]$^+$ calcd for $C_{23}H_{19}ClF_2N_3O_3S$, 490.08; found, 490.1.

***tert*-Butyl *N*-[4-[3-[2,6-difluoro-3-(propylsulfonylamino)benzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl]phenyl]carbamate (5)**

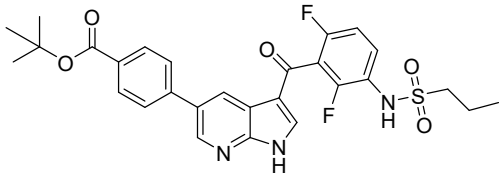


Compound **3** (458 mg, 1 mmol) and 4-(*N*-Boc-amino)phenylboronic acid (356 mg, 1.5 mmol) were suspended in dioxane (3 mL). After the addition of Na_2CO_3 solution (2M, 2 mL), the mixture was flushed with argon, and Pd(dppf)Cl_2 × CH_2Cl_2 (82 mg, 0.1 mmol) was added. It was heated *via* microwave

irradiation at 120 °C for 20 min. Subsequently, the mixture was poured onto H_2O (100 mL) and extracted with EtOAc (3 × 50 mL). The organic layers were washed with brine (50 mL), dried over Na_2SO_4, filtered and evaporated. The crude product was purified by flash chromatography (gradient of petroleum ether/EtOAc 1:1 to 1:2) to yield the title compound as a colourless solid.

Yield (364 mg, 64%); R_f = 0.40 (petroleum ether/EtOAc 1:1); mp 226 – 228 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.96 (t, *J* = 7.4 Hz, 3H), 1.50 (s, 9H), 1.74 (q, *J* = 7.6 Hz, 2H), 3.09 – 3.15 (m, 2H), 7.28 (t, *J* = 8.6 Hz, 1H), 7.54 – 7.69 (m, 5H), 8.19 (s, 1H), 8.56 (s, 1H), 8.67 (d, *J* = 2.2 Hz, 1H), 9.47 (s, 1H), 9.75 (s, 1H), 12.93 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.58, 16.81, 28.12, 53.50, 79.16, 112.30 (d, *J* = 22.8 Hz), 115.62, 117.51, 118.23 (t, *J* = 23.3 Hz), 118.67, 121.97 (d, *J* = 11.8 Hz), 126.35, 127.31, 128.73 (d, *J* = 9.2 Hz), 131.29, 131.66, 138.53, 139.20, 143.73, 148.58, 152.33 (dd, *J* = 8.7, 249.7 Hz), 152.76, 155.99 (dd, *J* = 6.7, 246.3 Hz), 180.58; **LC-MS** (ESI) t_R = 11.56 min, 94% purity, *m/z* $[M + H]^+$ calcd for $C_{28}H_{29}F_2N_4O_5S$, 571.18; found, 571.2; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{28}H_{29}F_2N_4O_5S$, 571.1821; found, 571.1811.

tert-Butyl 4-[3-[2,6-difluoro-3-(propylsulfonylamino)benzoyl]-1_H_-pyrrolo[2,3-_b_]pyridin-5-yl]benzoate (6)

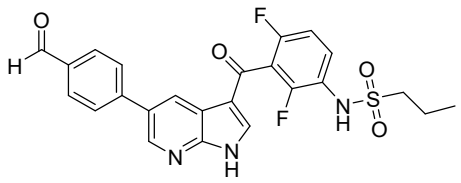


Compound **3** (1.15 g, 2.5 mmol), 4-(*tert*-butoxycarbonyl)phenylboronic acid (0.833 g, 3.75 mmol) and K_2CO_3 (0.691 g, 5 mmol) were suspended in dry toluene (20 mL). After the mixture was flushed with Argon, Pd(dppf)Cl_2 × CH_2Cl_2 (0.204 g, 0.25 mmol) was added. The reaction was stirred at 100 °C for 18 h, after which it was diluted with H_2O (100 mL) and extracted with EtOAc (3 × 100 mL). The combined organic layers were washed with brine (50 mL), dried over Na_2SO_4, filtered and concentrated *in vacuo*. After purification of the crude product by column chromatography (gradient of petroleum ether/EtOAc 2:1 to 1:1), the pale yellow solid was washed several times with EtOAc to obtain the title compound as a colourless solid.

Yield (0.62 g, 44%); R_f = 0.43 (petroleum ether/EtOAc 1:1); mp 228 – 230 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.97 (t, *J* = 7.4 Hz, 3H), 1.58 (s, 9H), 1.70 – 1.79 (m, 2H), 3.10 – 3.16 (m, 2H), 7.25 – 7.32 (m, 1H), 7.59 (td, *J* = 5.8, 9.0 Hz, 1H), 7.87 – 7.92 (m, 2H), 8.01 – 8.05 (m, 2H), 8.26 (s, 1H), 8.69 (s, 1H), 8.77 (d, *J* = 2.2 Hz, 1H), 9.75 (s, 1H), 13.05 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.58, 16.80, 27.81, 53.50, 80.79, 112.34 (d, *J* = 23.2 Hz), 115.74, 117.50, 117.83 – 118.69 (m), 121.56 – 122.48 (m), 127.20, 127.25 (d, *J* =

21.9 Hz), 128.82 (d, *J* = 9.3 Hz), 129.82, 130.28 (d, *J* = 10.9 Hz), 138.98, 142.37, 144.15, 149.23, 152.34 (dd, *J* = 8.6, 249.7 Hz), 156.00 (dd, *J* = 7.0, 246.4 Hz), 164.76, 180.67; **LC-MS** (ESI) t_R = 12.20 min, 98% purity, *m/z* [M + H]$^+$ calcd for $C_{28}H_{28}F_2N_3O_5S$, 556.17; found, 556.1; **HRMS** (ESI) *m/z* [M + H]$^+$ calcd for $C_{28}H_{28}F_2N_3O_5S$, 556.1712; found, 556.1712.

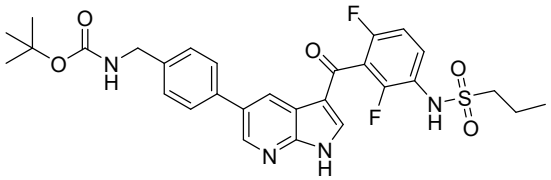
***N*-[2,4-Difluoro-3-[5-(4-formylphenyl)-1*H*-pyrrolo[2,3-*b*]pyridine-3-carbonyl]phenyl]propane-1-sulfonamide (7)**



Compound **3** (1.15 g, 2.5 mmol), 4-(formylphenyl)boronic acid (0.562 g, 3.75 mmol) and K_2CO_3 (0.691 g, 5 mmol) were suspended in DME/H_2O 4:1 (20 mL). After the mixture was flushed with argon, Pd(dppf)Cl_2 × CH_2Cl_2 (0.204 g, 0.25 mmol) was added. The reaction was stirred at 85 °C for 18 h, after which it was diluted with H_2O (100 mL) and extracted with EtOAc (3 × 100 mL). The combined organic layers were washed with brine (50 mL), dried over Na_2SO_4, filtered and concentrated *in vacuo*. After purification of the crude product by column chromatography (gradient of petroleum ether/EtOAc 2:1 to EtOAc), the pale yellow solid was washed several times with EtOAc to obtain the title product as a colourless solid.

Yield (0.81 g, 67%); R_f = 0.24 (petroleum ether/EtOAc 1:2); mp 260 – 262 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.97 (t, *J* = 7.4 Hz, 3H), 1.70 – 1.79 (m, 2H), 3.11 – 3.16 (m, 2H), 7.29 (td, *J* = 1.4, 8.8 Hz, 1H), 7.60 (td, *J* = 5.8, 9.0 Hz, 1H), 8.00 – 8.07 (m, 4H), 8.26 – 8.29 (m, 1H), 8.74 (s, 1H), 8.82 (d, *J* = 2.3 Hz, 1H), 9.75 (s, 1H), 10.09 (s, 1H), 13.05 – 13.08 (m, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 16.80, 53.49, 112.34 (d, *J* = 22.6 Hz), 115.78, 117.51, 118.09 (t, *J* = 23.3 Hz), 121.95 (d, *J* = 13.5 Hz), 127.54, 127.76, 128.83 (d, *J* = 9.9 Hz), 130.16, 130.28, 135.10, 139.07, 143.95, 144.27, 149.33, 152.34 (dd, *J* = 8.6, 249.8 Hz), 156.02 (dd, *J* = 6.6, 246.5 Hz), 180.67, 192.73; **LC-MS** (ESI) t_R = 10.87 min, 99% purity, *m/z* [M + H]$^+$ calcd for $C_{24}H_{20}F_2N_3O_4S$, 484.11; found, 484.3; **HRMS** (ESI) *m/z* [M + H]$^+$ calcd for $C_{24}H_{20}F_2N_3O_4S$, 484.1137; found, 484.1131.

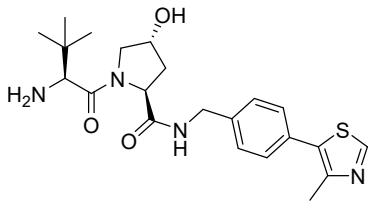
***tert*-Butyl *N*-[[4-[3-[2,6-difluoro-3-(propylsulfonylamino)benzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl] phenyl]methyl]carbamate (8)**[12]



The reductive amination was performed as described previously.[13] In detail, to a mixture of compound **7** (725 mg, 1.5 mmol) and *tert*-butyl carbamate (0.53 g, 4.5 mmol) in CH_2Cl_2/MeCN 1:3 (25 mL) were added Et_3SiH (0.72 mL, 4.5 mmol) and TFA (0.23 mL, 3 mmol) dropwise. After stirring at rt for 18 h, it was quenched with saturated $NaHCO_3$ solution (50 mL) and the aqueous layer was extracted with CH_2Cl_2 (3 × 50 mL). The combined organic layers were washed with H_2O (50 mL) and brine (50 mL). It was dried over Na_2SO_4, filtered and evaporated under reduced pressure. After purification by column chromatography (gradient of petroleum ether/EtOAc 1:1 to 1:2) and subsequent recrystallization from MeCN/EtOAc/MeOH 7:2:1 (10 mL), the title compound was obtained as a colourless solid.

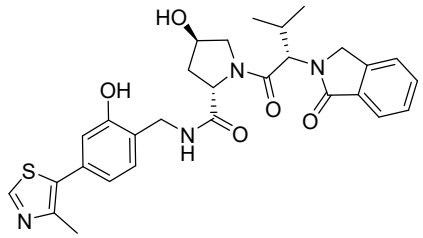
Yield (0.30 g, 34%); R_f = 0.20 (petroleum ether/EtOAc 1:2); mp 204 – 206 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.96 (t, *J* = 7.4 Hz, 3H), 1.41 (s, 9H), 1.70 – 1.79 (m, 2H), 3.10 – 3.16 (m, 2H), 4.20 (d, *J* = 6.2 Hz, 2H), 7.25 – 7.31 (m, 1H), 7.39 (d, *J* = 7.8 Hz, 2H), 7.44 (t, *J* = 6.3 Hz, 1H), 7.59 (td, *J* = 5.7, 9.0 Hz, 1H), 7.70 (d, *J* = 7.8 Hz, 2H), 8.22 (s, 1H), 8.61 (s, 1H), 8.70 (d, *J* = 2.4 Hz, 1H), 9.75 (s, 1H), 12.97 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 16.80, 28.25, 43.08, 53.49, 77.80, 112.00 – 112.78 (m), 115.65, 117.50, 118.19 (t, *J* = 23.5 Hz), 121.93 (d, *J* = 15.7 Hz), 126.81, 126.99, 127.75, 128.75 (d, *J* = 9.8 Hz), 131.35, 136.50, 138.66, 139.61, 143.95, 148.80, 152.32 (dd, *J* = 8.6, 249.7 Hz), 155.82, 156.00 (dd, *J* = 7.3, 246.5 Hz), 180.59; **LC-MS** (ESI) t_R = 11.10 min, 93% purity, *m*/*z* $[M + H]^+$ calcd for $C_{29}H_{31}F_2N_4O_5S$, 585.20; found, 585.4.

(2*S*,4*R*)-1-[(2*S*)-2-amino-3,3-dimethyl-butanoyl]-4-hydroxy-*N*-[[4-(4-methylthiazol-5-yl)phenyl]methyl] pyrrolidine-2-carboxamide (9)



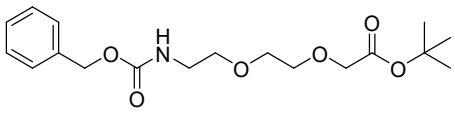
This compound was synthesized as we described previously.[13]

(2*S*,4*R*)-4-Hydroxy-*N*-[[2-hydroxy-4-(4-methylthiazol-5-yl)phenyl]methyl]-1-[(2*S*)-3-methyl-2-(1-oxoisoindolin-2-yl)butanoyl]pyrrolidine-2-carboxamide (10)



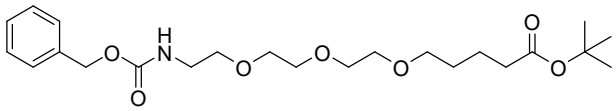
This compound was synthesized as we described previously.[13]

***tert*-Butyl 2-[2-[2-(benzyloxycarbonylamino)ethoxy]ethoxy]acetate (11a)**



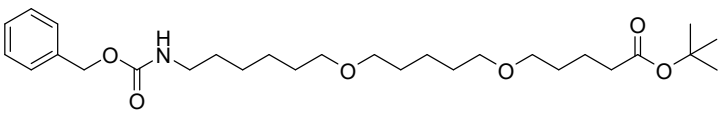
This compound was synthesized as we described previously.[9]

***tert*-Butyl 5-[2-[2-[2-(benzyloxycarbonylamino)ethoxy]ethoxy]ethoxy]pentanoate (11b)**



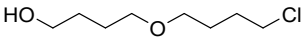
This compound was synthesized as we described previously.[9]

***tert*-Butyl 5-[5-[6-(benzyloxycarbonylamino)hexoxy]pentoxy]pentanoate (11c)**



This compound was synthesized as we described previously.[9]

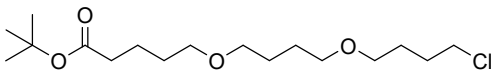
4-(4-Chlorobutoxy)butan-1-ol (29)[14]



Butane-1,4-diol (3.0 mL, 50 mmol) was suspended in DMSO (10 mL) and aqueous NaOH (19.4 M, 2.5 mL). After stirring for 10 minutes, 1-bromo-4-chlorobutane (1.7 mL, 10 mmol) was added while cooling with a water bath. The resulting mixture was stirred at rt for 18 h, after which it was acidified to *p*H 2 with 2N HCl. The product was extracted with CH_2Cl_2 (3 × 20 mL), washed with H_2O (20 mL) and brine (20 mL), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (gradient of petroleum ether/EtOAc 3:1 to 1:2) gave **29** as a colourless oil.

Yield (1.26 g, 70%); R_f = 0.54 (petroleum ether/EtOAc 1:2); **^{1}H NMR** (500 MHz, DMSO-d_6) δ 1.39 – 1.56 (m, 4H), 1.56 – 1.65 (m, 2H), 1.71 – 1.81 (m, 2H), 3.32 – 3.44 (m, 6H), 3.64 (t, *J* = 6.6 Hz, 2H), 4.33 (t, *J* = 5.1 Hz, 1H); **^{13}C NMR** (126 MHz, DMSO-d_6) δ 25.87, 26.58, 29.17, 29.25, 45.29, 60.53, 69.05, 69.90; **LC-MS** (ESI) t_R = 9.21 min, *m/z* $[M + H]^+$ calcd for $C_8H_{18}ClO_2$, 181.10; found, 181.0.

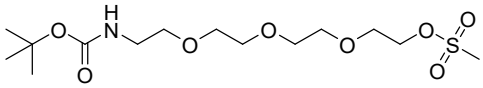
***tert*-Butyl 5-[4-(4-chlorobutoxy)butoxy]pentanoate (12a)**



tert-Butyl bromopentanoate (0.90 g, 5 mmol) and **29** (1.19 g, 5 mmol) were dissolved in toluene (5 mL). While cooling, TBAHS (1.70 g, 5 mmol) and 50% aqueous NaOH solution (2.5 mL) were added. The reaction mixture was stirred at rt for 16 h. Subsequently it was diluted with H_2O (30 mL) and extracted with EtOAc (3 × 30 mL). The combined organic layers were washed with H_2O (30 mL) and brine (30 mL), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Purification by column chromatography (gradient of petroleum ether/EtOAc 10:1 to 8:1) gave the title compound as a colourless oil.

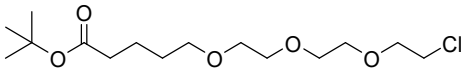
Yield (175 mg, 10%); R_f = 0.40 (petroleum ether/EtOAc 8:1); **^{1}H NMR** (600 MHz, $CDCl_3$) δ 1.43 (s, 9H), 1.54 – 1.75 (m, 10H), 1.81 – 1.89 (m, 2H), 2.23 (t, *J* = 7.3 Hz, 2H), 3.37 – 3.48 (m, 8H), 3.56 (t, *J* = 6.7 Hz, 2H); **^{13}C NMR** (151 MHz, $CDCl_3$) δ 22.00, 26.62, 27.24, 28.25, 29.28, 29.70, 35.45, 45.14, 69.99, 70.54, 70.77, 70.82, 80.16, 173.17; **LC-MS** (ESI) t_R = 12.48 min, *m/z* $[M + H]^+$ calcd for $C_{17}H_{34}ClO_4$, 337.21; found, 337.1; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{17}H_{34}ClO_4$, 337.2140; found, 337.2132.

2-[2-[2-[2-(*tert*-Butoxycarbonylamino)ethoxy]ethoxy]ethoxy]ethyl methanesulfonate (12b)



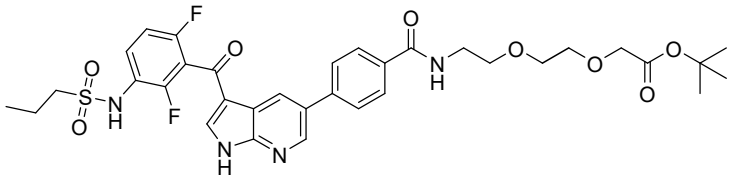
This compound was synthesized as we described previously.[15]

***tert*-Butyl 5-[2-[2-(2-chloroethoxy)ethoxy]ethoxy]pentanoate (12c)**



This compound was synthesized as we described previously.[9]

***tert*-Butyl 2-[2-[2-[[4-[3-[2,6-difluoro-3-(propylsulfonylamino)benzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl]benzoyl]amino]ethoxy]ethoxy]acetate (13a)**

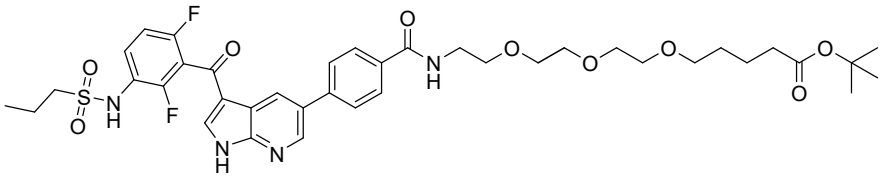


Linker **11a** (88 mg, 0.25 mmol) was dissolved in dry EtOAc (2.5 mL) and 10% Pd/C (10 mg) was added. The mixture was stirred under a hydrogen atmosphere at rt for 18 h. The suspension was filtered, and the filtrate was concentrated *in vacuo*. In a separate flask, vemurafenib derivative **6** (0.14 g, 0.25 mmol) was dissolved in dry CH_2Cl_2/TFA 4:1 (5 mL) and stirred at rt for 4 h. After removal of the volatiles under reduced pressure, it was co-evaporated with dry CH_2Cl_2 (2 × 5 mL) and further dried under high vacuum. The deprotected acid was dissolved in dry DMF (2.5 mL) and treated with DIPEA (0.26 mL, 1.5 mmol) and HATU (0.10 g, 0.275 mmol). After stirring for 5 min at rt, the deprotected amine in dry DMF (2.5 mL) was added and the mixture was stirred at rt for 16 h. The mixture was diluted with H_2O (50 mL) and extracted with EtOAc (3 × 25 mL). The combined organic layers were washed with brine (25 mL), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Purification by column chromatography (gradient of CH_2Cl_2/MeOH 39:1 to 19:1) gave the title compound as a colourless solid.

Yield (86 mg, 49%); R_f = 0.38 (CH_2Cl_2/MeOH 19:1); mp 94 – 96 °C; **^{1}H NMR** (600 MHz, $CDCl_3$) δ 0.96 (t, *J* = 7.4 Hz, 3H), 1.40 (s, 9H), 1.70 – 1.79 (m, 2H), 3.10 – 3.15 (m, 2H), 3.44 – 3.49 (m, 2H), 3.55 – 3.64 (m, 6H),

3.99 (s, 2H), 7.26 – 7.32 (m, 1H), 7.59 (td, *J* = 5.8, 9.0 Hz, 1H), 7.86 (d, *J* = 8.1 Hz, 2H), 7.98 – 8.03 (m, 2H), 8.26 (d, *J* = 3.1 Hz, 1H), 8.61 (t, *J* = 5.6 Hz, 1H), 8.69 (s, 1H), 8.77 (d, *J* = 2.2 Hz, 1H), 9.78 (s, 1H), 13.05 (s, 1H); **^{13}C NMR** (151 MHz, $CDCl_3$) δ 12.62, 16.84, 27.76, 53.46, 68.13, 68.90, 69.55, 69.87, 80.66, 112.39 (d, *J* = 23.0 Hz), 115.74, 117.53, 118.15 (t, *J* = 23.4 Hz), 121.95 (d, *J* = 11.8 Hz), 126.94, 127.24, 128.09, 128.84, 130.60, 133.32, 139.01, 140.72, 144.15, 149.13, 151.28 – 153.62 (m), 154.92 – 157.27 (m), 165.90, 169.39, 180.70; **LC-MS** (ESI) t_R = 11.07 min, 95% purity, *m*/*z* $[M + H]^+$ calcd for $C_{34}H_{39}F_2N_4O_8S$, 701.25; found, 701.5; **HRMS** (ESI) *m*/*z* $[M + H]^+$ calcd for $C_{34}H_{39}F_2N_4O_8S$, 701.2451; found, 701.2434.

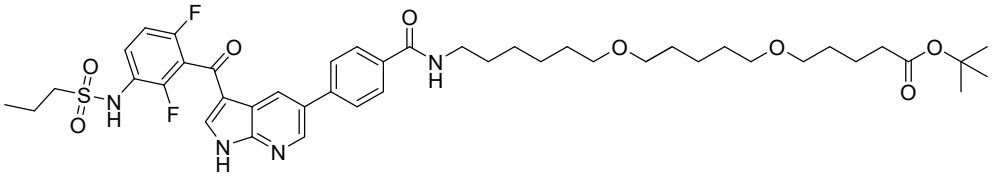
***tert*-Butyl 5-[2-[2-[2-[[4-[3-[2,6-difluoro-3-(propylsulfonylamino)benzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl]benzoyl]amino]ethoxy]ethoxy]ethoxy]pentanoate (13b)**



This compound was synthesized by analogy with **13a** but using linker **11b** (110 mg, 0.25 mmol). The crude product was purified by column chromatography silica gel (gradient from CH_2Cl_2/MeOH 39:1 to 19:1) to yield the title compound as a colourless solid.

Yield (121 mg, 61%); R_f = 0.31 (CH_2Cl_2/MeOH 15:1); mp 80 – 82 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.97 (t, *J* = 7.5 Hz, 3H), 1.36 (s, 9H), 1.42 – 1.53 (m, 4H), 1.70 – 1.79 (m, 2H), 2.16 (t, *J* = 7.0 Hz, 2H), 3.10 – 3.16 (m, 2H), 3.35 (t, *J* = 6.1 Hz, 2H), 3.42 – 3.49 (m, 4H), 3.49 – 3.60 (m, 8H), 7.25 – 7.32 (m, 1H), 7.54 – 7.63 (m, 1H), 7.84 – 7.89 (m, 2H), 7.98 – 8.03 (m, 2H), 8.25 (s, 1H), 8.58 (t, *J* = 5.6 Hz, 1H), 8.69 (s, 1H), 8.77 (d, *J* = 2.3 Hz, 1H), 9.74 (s, 1H), 13.02 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 16.80, 21.41, 27.71, 28.43, 34.44, 53.49, 68.89, 69.42, 69.64, 69.75, 69.77, 69.83, 78.90 – 79.80 (m), 112.31 (dd, *J* = 3.5, 22.9 Hz), 115.72, 117.49, 117.87 – 118.82 (m), 121.95 (d, *J* = 13.4 Hz), 126.87, 127.19, 128.03, 128.78, 130.56, 133.30, 138.87, 140.68, 144.10, 149.10, 152.33 (dd, *J* = 8.7, 249.7 Hz), 156.00 (dd, *J* = 6.6, 246.5 Hz), 165.83, 172.17, 180.63; **LC-MS** (ESI) t_R = 11.61 min, 96% purity, *m*/*z* $[M + H]^+$ calcd for $C_{39}H_{49}F_2N_4O_9S$, 787.32; found, 787.5; **HRMS** (ESI) *m*/*z* $[M + H]^+$ calcd for $C_{39}H_{49}F_2N_4O_9S$, 787.3183; found, 787.3161.

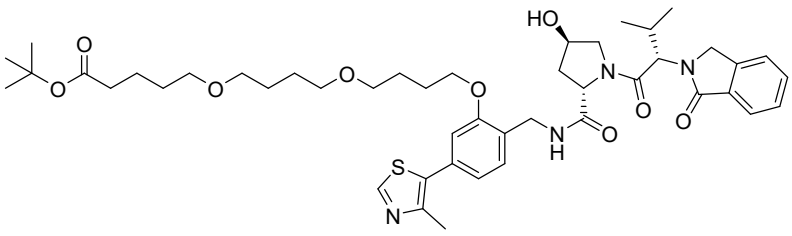
***tert*-Butyl 5-[5-[6-[[4-[3-[2,6-difluoro-3-(propylsulfonylamino)benzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl]benzoyl]amino]hexoxy]pentoxy]pentanoate (13c)**



This compound was synthesized by analogy with **13a** but using linker **11c** (123 mg, 0.25 mmol). The crude product was purified by column chromatography silica gel (CH_2Cl_2/MeOH 39:1) to yield the title compound as a colourless solid.

Yield (95 mg, 45%); R_f = 0.35 (CH_2Cl_2/MeOH 19:1); mp 88 – 90 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.97 (t, *J* = 7.5 Hz, 3H), 1.26 – 1.36 (m, 6H), 1.37 (s, 9H), 1.43 – 1.57 (m, 12H), 1.70 – 1.79 (m, 2H), 2.17 (t, *J* = 7.0 Hz, 2H), 3.13 (dd, *J* = 6.4, 8.9 Hz, 2H), 3.25 – 3.36 (m, 10H), 7.28 (t, *J* = 8.7 Hz, 1H), 7.56 – 7.63 (m, 1H), 7.85 (d, *J* = 8.0 Hz, 2H), 7.99 (d, *J* = 8.0 Hz, 2H), 8.25 (s, 1H), 8.51 (t, *J* = 5.6 Hz, 1H), 8.69 (s, 1H), 8.77 (d, *J* = 2.2 Hz, 1H), 9.75 (s, 1H), 13.02 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 16.80, 21.51, 22.49, 25.49, 26.33, 27.72, 28.48, 29.01, 29.02, 29.08, 29.18, 34.49, 53.50, 69.46, 69.82, 69.88, 79.30, 112.31 (d, *J* = 22.3 Hz), 115.72, 117.50, 117.92 – 118.62 (m), 121.96 (d, *J* = 13.5 Hz), 126.85, 127.17, 127.99, 128.79 (d, *J* = 9.6 Hz), 130.61, 133.61, 138.87, 140.55, 144.10, 149.10, 152.34 (dd, *J* = 8.4, 249.5 Hz), 156.01 (dd, *J* = 7.0, 246.7 Hz), 165.63, 172.18, 180.64; **LC-MS** (ESI) t_R = 12.21 min, 92% purity, *m/z* $[M + H]^+$ calcd for $C_{44}H_{59}F_2N_4O_8S$, 841.40; found, 841.5; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{44}H_{59}F_2N_4O_8S$, 841.4016; found, 841.3998.

***tert*-Butyl 5-[4-[4-[2-[[[(2*S*,4*R*)-4-hydroxy-1-[(2*S*)-3-methyl-2-(1-oxoisoindolin-2-yl)butanoyl]-pyrrolidine-2-carbonyl]amino]methyl]-5-(4-methylthiazol-5-yl)phenoxy]butoxy]butoxy]pentanoate (14a)**

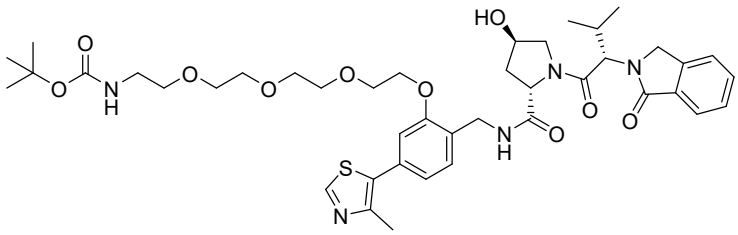


VHL ligand **10** (274 mg, 0.5 mmol) and linker **12a** (202 mg, 0.6 mmol) were dissolved in DMF (10 mL). K_2CO_3 (173 mg, 1.25 mmol) was added, and the mixture was heated to 70 °C for 18 h. It was quenched

with half-saturated brine (20 mL) and extracted with EtOAc (3 × 20 mL). The combined organic layers were washed with 5% LiCl solution and brine (20 mL each), dried over Na_2SO_4, filtrated and concentrated *in vacuo*. Purification by column chromatography (CH_2Cl_2/MeOH 19:1) yielded the title compound as a yellow oil.

Yield (164 mg, 39%); R_f = 0.31 (CH_2Cl_2/MeOH 19:1); **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.74 (d, *J* = 6.7 Hz, 3H), 0.96 (d, *J* = 6.5 Hz, 3H), 1.38 (s, 9H), 1.44 – 1.53 (m, 8H), 1.65 – 1.72 (m, 2H), 1.75 – 1.84 (m, 2H), 1.93 (ddd, *J* = 4.8, 8.0, 12.7 Hz, 1H), 2.00 – 2.07 (m, 1H), 2.18 (t, *J* = 7.1 Hz, 2H), 2.28 – 2.36 (m, 1H), 2.47 (s, 3H), 3.27 – 3.47 (m, 8H), 3.66 – 3.71 (m, 1H), 3.78 (dd, *J* = 4.5, 10.6 Hz, 1H), 4.07 (t, *J* = 6.3 Hz, 2H), 4.23 (dd, *J* = 5.9, 16.2 Hz, 1H), 4.27 – 4.59 (m, 5H), 4.71 (d, *J* = 10.8 Hz, 1H), 5.07 (d, *J* = 4.2 Hz, 1H), 6.97 – 7.02 (m, 2H), 7.34 (d, *J* = 7.6 Hz, 1H), 7.46 – 7.53 (m, 1H), 7.57 – 7.63 (m, 1H), 7.62 (s, 1H), 7.71 (d, *J* = 7.5 Hz, 1H), 8.34 (t, *J* = 6.0 Hz, 1H), 8.98 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 15.96, 18.58, 18.85, 21.51, 25.68, 25.86, 26.05, 26.08, 27.73, 28.35, 28.47, 34.48, 37.00, 38.04, 46.78, 55.36, 57.76, 58.66, 67.56, 68.58, 69.44, 69.60, 69.68, 69.74, 79.31, 111.68, 120.74, 122.98, 123.57, 126.96, 127.70, 127.86, 130.95, 131.27, 131.36, 131.54, 142.17, 147.85, 151.38, 155.89, 167.44, 168.08, 171.48, 172.18; **LC-MS** (ESI) t_R = 12.21 min, 96% purity, *m/z* $[M + H]^+$ calcd for $C_{46}H_{65}N_4O_9S$, 849.45; found, 849.5; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{46}H_{65}N_4O_9S$, 849.4467; found, 849.4450.

tert-Butyl _N_-[2-[2-[2-[2-[2-[[[(2_S_,4_R_)-4-hydroxy-1-[(2_S_)-3-methyl-2-(1-oxoisoindolin-2-yl)butanoyl]pyrrolidine-2-carbonyl]amino]methyl]-5-(4-methylthiazol-5-yl)phenoxy]ethoxy]ethoxy]ethoxy]ethyl]carbamate (14b)

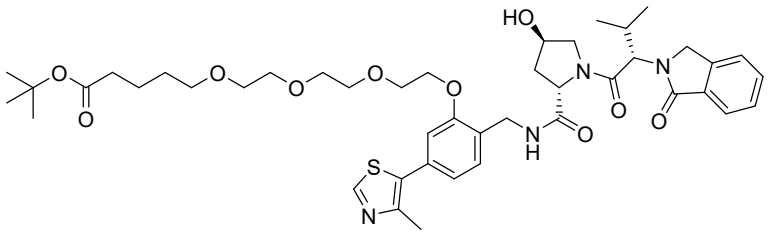


This compound was synthesized by analogy with **14a** but using linker **12b** (223 mg, 0.25 mmol). The crude product was purified by column chromatography silica gel (CH_2Cl_2/MeOH 19:1) to yield the title compound as a colourless solid.

Yield (360 mg, 87%); R_f = 0.31 (CH_2Cl_2/MeOH 15:1); mp 62 – 64 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.74 (d, *J* = 6.6 Hz, 3H), 0.97 (d, *J* = 6.5 Hz, 3H), 1.36 (s, 9H), 1.89 – 1.96 (m, 1H), 2.01 – 2.08 (m, 1H), 2.28 – 2.38 (m, 1H), 2.47 (s, 3H), 3.05 (q, *J* = 6.0 Hz, 2H), 3.36 (t, *J* = 6.1 Hz, 2H), 3.47 (dd, *J* = 3.6, 6.1 Hz, 2H), 3.51 (dd,

J = 3.6, 6.0 Hz, 2H), 3.55 (dd, *J* = 3.7, 5.9 Hz, 2H), 3.63 (dd, *J* = 3.7, 5.8 Hz, 2H), 3.66 – 3.72 (m, 1H), 3.74 – 3.84 (m, 3H), 4.19 (dd, *J* = 3.7, 5.7 Hz, 2H), 4.21 – 4.60 (m, 6H), 4.72 (d, *J* = 10.8 Hz, 1H), 5.07 (d, *J* = 4.1 Hz, 1H), 6.71 (t, *J* = 5.8 Hz, 1H), 7.01 (dd, *J* = 1.6, 7.8 Hz, 1H), 7.05 (d, *J* = 1.6 Hz, 1H), 7.34 (d, *J* = 7.8 Hz, 1H), 7.50 (ddd, *J* = 2.2, 6.1, 8.0 Hz, 1H), 7.57 – 7.65 (m, 2H), 7.71 (d, *J* = 7.6 Hz, 1H), 8.34 (t, *J* = 5.9 Hz, 1H), 8.98 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 15.98, 18.59, 18.85, 28.19, 28.35, 37.03, 38.06, 40.06, 46.78, 55.37, 57.76, 58.68, 67.90, 68.59, 68.98, 69.13, 69.48, 69.74, 69.82, 70.05, 77.53, 112.18, 121.06, 122.98, 123.58, 127.18, 127.69, 127.87, 130.96, 131.22, 131.36, 131.54, 142.17, 147.91, 151.41, 155.55, 155.86, 167.45, 168.08, 171.51; **LC-MS** (ESI) t_R = 11.12 min, 98% purity, *m*/*z* [M + H]$^+$ calcd for $C_{42}H_{58}N_5O_{10}S$, 824.39; found, 824.6; **HRMS** (ESI) *m*/*z* [M + H]$^+$ calcd for $C_{42}H_{58}N_5O_{10}S$, 824.3899; found, 824.3878.

tert-Butyl 5-[2-[2-[2-[2-[[[(2*S*,4*R*)-4-hydroxy-1-[(2*S*)-3-methyl-2-(1-oxoisoindolin-2-yl)butanoyl]pyrrolidine-2-carbonyl]amino]methyl]-5-(4-methylthiazol-5-yl)phenoxy]ethoxy]ethoxy]ethoxy]pentanoate (14c)

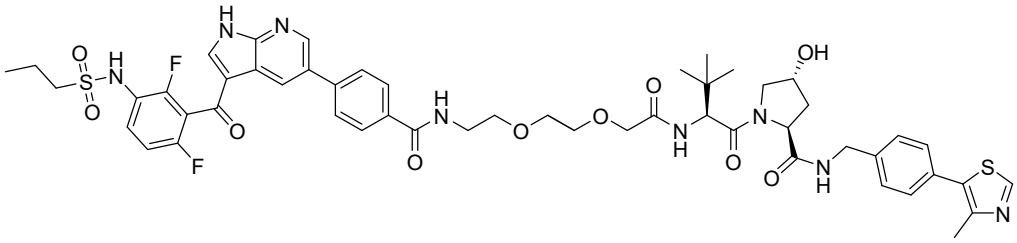


This compound was synthesized by analogy with **14a** but using linker **12c** (195 mg, 0.25 mmol). The crude product was purified by column chromatography silica gel (CH_2Cl_2/MeOH 29:1) to yield the title compound as a yellow oil.

Yield (106 mg, 25%); R_f = 0.43 (CH_2Cl_2/MeOH 15:1); **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.74 (d, *J* = 6.7 Hz, 3H), 0.97 (d, *J* = 6.4 Hz, 3H), 1.37 (s, 9H), 1.43 – 1.52 (m, 4H), 1.92 (ddd, *J* = 4.7, 8.0, 12.7 Hz, 1H), 2.04 (ddd, *J* = 2.5, 7.6, 11.8 Hz, 1H), 2.17 (t, *J* = 7.0 Hz, 2H), 2.32 (dq, *J* = 6.5, 10.8 Hz, 1H), 2.47 (s, 3H), 3.35 (t, *J* = 6.0 Hz, 2H), 3.44 (dd, *J* = 3.8, 6.0 Hz, 2H), 3.51 (dd, *J* = 3.9, 5.8 Hz, 2H), 3.55 (dd, *J* = 3.7, 5.9 Hz, 2H), 3.63 (dd, *J* = 3.8, 5.9 Hz, 2H), 3.66 – 3.71 (m, 1H), 3.75 – 3.82 (m, 3H), 4.17 – 4.21 (m, 2H), 4.22 – 4.31 (m, 2H) 4.33 (td, *J* = 3.6, 6.1 Hz, 1H), 4.41 (t, *J* = 7.9 Hz, 1H), 4.46 (d, *J* = 18.0 Hz, 1H), 4.55 (d, *J* = 18.1 Hz, 1H), 4.71 (d, *J* = 10.8 Hz, 1H), 7.01 (dd, *J* = 1.6, 7.7 Hz, 1H), 7.05 (d, *J* = 1.7 Hz, 1H), 7.34 (d, *J* = 7.8 Hz, 1H), 7.50 (ddd, *J* = 2.3, 6.0, 7.9 Hz), 7.65 – 7.57 (m, 2H), 7.71 (d, *J* = 7.6 Hz, 1H), 8.33 (t, *J* = 6.0 Hz, 1H), 8.99 (s, 1H). The signal for OH is missing. **^{13}C NMR** (151 MHz, DMSO-d_6) δ 15.97, 18.59, 18.85, 21.43, 27.73, 28.35, 28.44, 34.46, 37.03, 38.05, 46.78, 55.37, 57.76, 58.68, 67.90, 68.59, 68.98, 69.41, 69.78, 69.82, 69.85, 70.06, 79.33, 112.17, 121.05, 122.98, 123.58, 127.20, 127.70, 127.87, 130.95, 131.23, 131.36, 131.54, 142.17, 147.88,

151.42, 155.86, 167.45, 168.07, 171.50, 172.19; **LC-MS** (ESI) t_R = 11.64 min, 96% purity, *m*/*z* $[M + H]^+$ calcd for $C_{44}H_{61}N_4O_{10}S$, 837.41; found, 837.9; **HRMS** (ESI) *m*/*z* $[M + H]^+$ calcd for $C_{44}H_{61}N_4O_{10}S$, 837.4103; found, 837.4081.

BRAF-targeting PROTAC 15a

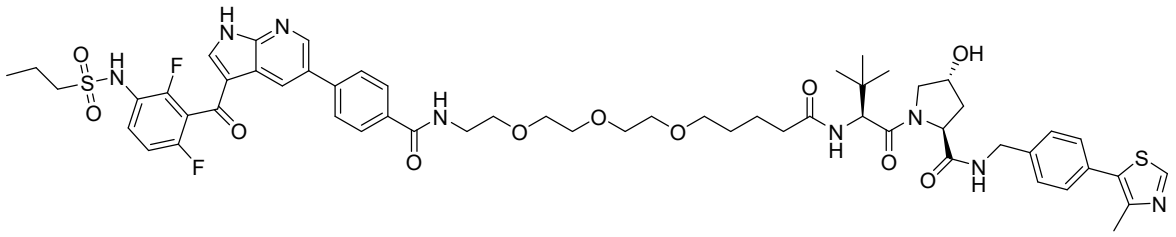


Conjugate **13a** (70 mg, 0.1 mmol) and VHL ligand **9** (53 mg, 0.1 mmol) were threated in separate flasks with TFA/dry CH_2Cl_2 4:1 (5 mL) at rt for 4 h. After removal of the volatiles, the residue was co-evaporated with dry CH_2Cl_2 (2 × 5 mL), and the material was dried under high vacuum. After reconstitution with dry DMF (each 5 mL) and addition of DIPEA (each 39 mg, 52 µL, 0.3 mmol), HATU (42 mg, 0.11 mmol) was added to the first flask containing the free acid derived from **13a**. After stirring for 5 min at rt, both solutions were combined, and it was stirred at rt for 18 h. Subsequently, it was diluted with H_2O (50 mL), extracted with EtOAc (3 × 25 mL), and the combined organic layers were washed with 5% LiCl solution and brine (each 50 mL). The solution was dried over Na_2SO_4, filtered, and concentrated *in vacuo*. The crude product was purified by column chromatography silica gel (gradient from CH_2Cl_2/MeOH 19:1 to 9:1) to yield the title compound as a colourless solid.

Yield (35 mg, 33%); R_f = 0.14 (CH_2Cl_2/MeOH 15:1); mp 176 – 178 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.91 – 0.98 (m, 12H), 1.70 – 1.78 (m, 2H), 1.81 – 2.00 (m, 2H), 2.04 – 2.10 (m, 1H), 2.43 (s, 3H), 3.10 – 3.15 (m, 2H), 3.48 (q, *J* = 5.8 Hz, 2H), 3.57 – 3.71 (m, 8H), 3.98 (s, 1H), 4.25 (dd, *J* = 5.8, 15.7 Hz, 1H), 4.37 (dd, *J* = 6.2, 15.7 Hz, 2H), 4.47 (t, *J* = 8.1 Hz, 1H), 4.58 (d, *J* = 9.5 Hz, 1H), 5.18 (d, *J* = 3.6 Hz, 1H), 7.28 (t, *J* = 8.9 Hz, 1H), 7.36 – 7.47 (m, 5H), 7.55 – 7.63 (m, 1H), 7.82 – 7.87 (m, 2H), 7.99 (d, *J* = 8.4 Hz, 2H), 8.23 (s, 1H), 8.54 – 8.65 (m, 2H), 8.68 (s, 1H), 8.76 (d, *J* = 2.3 Hz, 1H), 8.95 (s, 1H), 9.74 (s, 1H), 13.07 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.59, 15.89, 16.81, 26.19, 35.72, 37.92, 41.68, 53.50, 55.73, 56.57, 58.73, 68.84, 69.04, 69.38, 69.57, 70.41, 112.30 (d, *J* = 23.2 Hz), 115.71, 117.51, 118.13 (t, *J* = 23.5 Hz), 121.83 – 122.26 (m), 126.89, 127.20, 127.45, 128.07, 128.80, 129.70, 130.58, 131.11, 133.27, 138.90, 139.39, 140.68, 144.11, 147.72, 149.09, 151.38, 153.20 (d, *J* = 8.9 Hz), 155.95 (d, *J* = 246.4 Hz), 165.92, 168.63, 169.20,

171.69, 180.67; **LC-MS** (ESI) t_R = 11.16 min, 96% purity, *m/z* [M + H]$^+$ calcd for $C_{52}H_{59}F_2N_8O_{10}S_2$, 1057.38; found, 1057.6; **HRMS** (ESI) *m/z* [M + H]$^+$ calcd for $C_{52}H_{59}F_2N_8O_{10}S_2$, 1057.3758; found, 1057.3760.

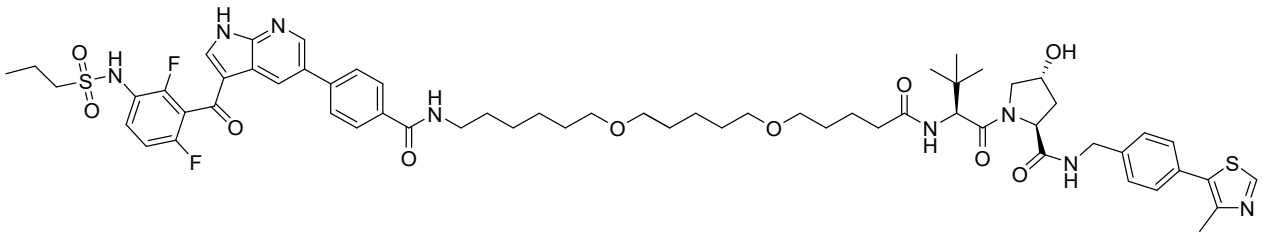
BRAF-targeting PROTAC 15b



This compound was synthesized by analogy with **15a** but using conjugate **13b** (79 mg, 0.1 mmol). The crude product was purified by column chromatography silica gel (gradient from CH_2Cl_2/MeOH 19:1 to 9:1), followed by preparative HPLC (H_2O/MeOH 25/75) to yield the title compound as a colourless solid.

Yield (69 mg, 61%); R_f = 0.14 (CH_2Cl_2/MeOH 15:1); mp 144 – 146 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.93 (s, 9H), 0.96 (t, *J* = 7.4 Hz, 3H), 1.42 – 1.55 (m, 4H), 1.71 – 1.78 (m, 2H), 1.87 – 1.93 (m, 1H), 2.00 – 2.05 (m, 1H), 2.10 – 2.16 (m, 1H), 2.22 – 2.30 (m, 1H), 2.44 (s, 3H), 3.10 – 3.14 (m, 2H), 3.36 (t, *J* = 6.1 Hz, 2H), 3.43 – 3.47 (m, 4H), 3.50 – 3.57 (m, 8H), 3.61 – 3.69 (m, 2H), 4.21 (dd, *J* = 5.5, 15.8 Hz, 1H), 4.32 – 4.37 (m, 1H), 4.39 – 4.46 (m, 2H), 4.54 (d, *J* = 9.2 Hz, 1H), 5.10 (d, *J* = 3.6 Hz, 1H), 7.25 – 7.30 (m, 1H), 7.36 – 7.43 (m, 4H), 7.55 – 7.62 (m, 1H), 7.84 (dd, *J* = 8.8, 25.7 Hz, 3H), 7.97 – 8.02 (m, 2H), 8.24 (s, 1H), 8.53 (t, *J* = 6.1 Hz, 1H), 8.59 (t, *J* = 5.6 Hz, 1H), 8.69 (s, 1H), 8.77 (d, *J* = 2.2 Hz, 1H), 8.97 (s, 1H), 9.75 (s, 1H), 13.01 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.58, 15.90, 16.81, 22.18, 26.34, 28.74, 34.58, 35.16, 37.91, 41.63, 53.48, 56.28, 56.31, 58.66, 68.84, 68.89, 69.43, 69.62, 69.73, 69.76, 70.00, 112.29 (d, *J* = 21.8 Hz), 115.72, 117.49, 117.90 – 118.34 (m), 121.97 – 122.18 (m), 126.88, 127.18, 128.60 – 128.80 (m), 127.40, 128.04, 128.60, 128.45 – 129.12 (m), 129.61, 130.55, 131.13, 133.29, 138.88, 139.47, 140.68, 144.10, 147.69, 149.10, 151.38, 151.44 – 153.36 (m), 154.44 – 158.22 (m), 165.86, 169.69, 171.90, 171.95, 180.66; **LC-MS** (ESI) t_R = 11.08 min, 99% purity, *m/z* [M + H]$^+$ calcd for $C_{57}H_{69}F_2N_8O_{11}S_2$, 1143.45; found, 1144.0; **HRMS** (ESI) *m/z* [M + H]$^+$ calcd for $C_{57}H_{69}F_2N_8O_{11}S_2$, 1143.4490; found, 1143.4495.

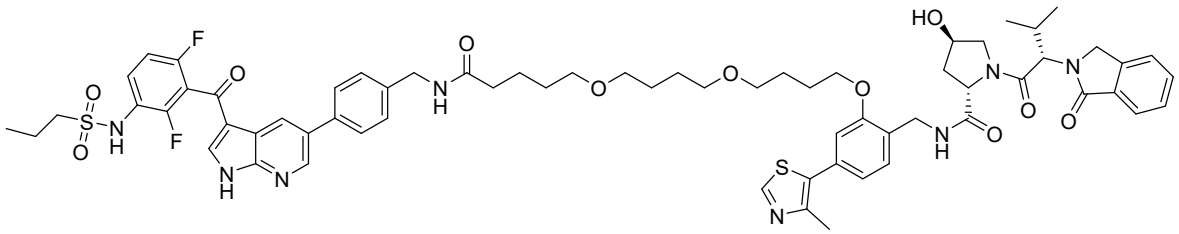
BRAF-targeting PROTAC 15c



This compound was synthesized by analogy with **15a** but using conjugate **13c** (84 mg, 0.1 mmol). The crude product was purified by column chromatography silica gel (gradient from CH_2Cl_2/MeOH 19:1 to 9:1), followed by preparative HPLC (H_2O/MeOH 20/80) to yield the title compound as a colourless solid.

Yield (26 mg, 22%); R_f = 0.22 (CH_2Cl_2/MeOH 15:1); mp 110 – 112 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.93 (s, 9H), 0.96 (t, *J* = 7.4 Hz, 3H), 1.27 – 1.35 (m, 6H), 1.43 – 1.56 (m, 12H), 1.70 – 1.79 (m, 2H), 1.86 – 1.94 (m, 1H), 1.99 – 2.06 (m, 1H), 2.07 – 2.16 (m, 1H), 2.22 – 2.30 (m, 1H), 2.44 (s, 3H), 3.11 – 3.15 (m, 2H), 3.26 – 3.31 (m, 8H), 3.34 (d, *J* = 6.7 Hz, 2H), 3.61 – 3.70 (m, 2H), 4.21 (dd, *J* = 5.5, 15.8 Hz, 1H), 4.32 – 4.37 (m, 1H), 4.39 – 4.46 (m, 2H), 4.54 (d, *J* = 9.4 Hz, 1H), 5.10 (d, *J* = 3.6 Hz, 1H), 7.28 (t, *J* = 8.8 Hz, 1H), 7.35 – 7.44 (m, 4H), 7.55 – 7.63 (m, 1H), 7.83 (dd, *J* = 8.6, 19.6 Hz, 3H), 7.96 – 8.01 (m, 2H), 8.25 (s, 1H), 8.48 – 8.56 (m, 2H), 8.68 (s, 1H), 8.77 (d, *J* = 2.2 Hz, 1H), 8.97 (s, 1H), 9.75 (s, 1H), 13.02 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 15.90, 16.79, 22.24, 22.48, 25.48, 26.34, 28.79, 29.03, 29.08, 29.18, 34.60, 35.17, 37.91, 41.63, 53.48, 56.27, 56.30, 58.66, 68.83, 69.61, 69.85, 69.88, 112.32 (d, *J* = 21.6 Hz), 115.71, 117.48, 117.81 – 118.50 (m), 121.71 – 122.19 (m), 126.85, 127.16, 127.40, 127.98, 128.60, 128.66 – 129.03 (m), 129.61, 130.60, 131.13, 133.59, 138.87, 139.47, 140.53, 144.10, 147.69, 149.08, 151.38, 151.43 – 153.32 (m), 154.98 – 157.13 (m), 165.62, 169.68, 171.90, 171.95, 180.63; **LC-MS** (ESI) t_R = 11.91 min, 99% purity, *m/z* $[M + H]^+$ calcd for $C_{62}H_{79}F_2N_8O_{10}S_2$, 1197.53; found, 1198.2; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{62}H_{79}F_2N_8O_{10}S_2$, 1197.5323; found, 1197.5326.

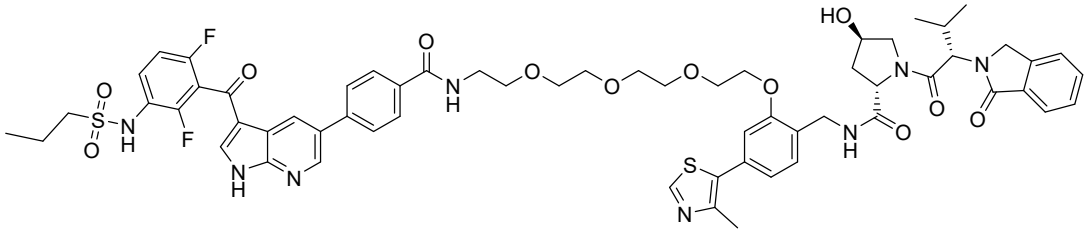
BRAF-targeting PROTAC 16a



Conjugate **14a** (85 mg, 0.1 mmol) and BRAF ligand **8** (58 mg, 0.1 mmol) were threated in separate flasks with TFA/dry CH_2Cl_2 4:1 (5 mL) at rt for 4 h. After removal of the volatiles, the residue was co-evaporated with dry CH_2Cl_2 (2 × 5 mL), and the material was dried under high vacuum. After reconstitution with dry DMF (each 5 mL) and addition of DIPEA (each 39 mg, 52 µL, 0.3 mmol), HATU (42 mg, 0.11 mmol) was added to the first flask containing the free acid derived from **14a**. After stirring for 5 min at rt, both solutions were combined, and it was stirred at rt for 18 h. Subsequently, it was diluted with H_2O (50 mL), extracted with EtOAc (3 × 25 mL), and the combined organic layers were washed with 5% LiCl solution and brine (each 50 mL). The solution was dried over Na_2SO_4, filtered, and concentrated *in vacuo*. The crude product was purified by column chromatography silica gel (gradient from CH_2Cl_2/MeOH 19:1 to 9:1) followed by preparative HPLC (H_2O/MeOH 20/80) to yield the title compound as a colourless solid.

Yield (83 mg, 66%); R_f = 0.21 (CH_2Cl_2/MeOH 15:1); mp 134 – 136 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.73 (d, *J* = 6.6 Hz, 3H), 0.93 – 0.99 (m, 6H), 1.46 – 1.53 (m, 6H), 1.55 – 1.61 (m, 2H), 1.64 – 1.70 (m, 2H), 1.71 – 1.82 (m, 4H), 1.89 – 1.96 (m, 1H), 2.00 – 2.09 (m, 1H), 2.17 (t, *J* = 7.3 Hz, 2H), 2.26 – 2.37 (m, 1H), 2.46 (s, 3H), 3.09 – 3.15 (m, 2H), 3.27 – 3.40 (m, 6H), 3.41 (t, *J* = 6.4 Hz, 2H), 3.66 – 3.71 (m, 1H), 3.77 (dd, *J* = 4.5, 10.6 Hz, 1H), 4.06 (t, *J* = 6.3 Hz, 2H), 4.23 (dd, *J* = 5.8, 16.3 Hz, 1H), 4.31 (dd, *J* = 5.9, 19.1 Hz, 4H), 4.38 – 4.49 (m, 2H), 4.55 (d, *J* = 18.0 Hz, 1H), 4.71 (d, *J* = 10.8 Hz, 1H), 5.07 (d, *J* = 4.1 Hz, 1H), 6.99 (d, *J* = 7.8 Hz, 2H), 7.24 – 7.31 (m, 1H), 7.33 (d, *J* = 7.6 Hz, 1H), 7.36 – 7.42 (m, 2H), 7.43 – 7.53 (m, 1H), 7.55 – 7.64 (m, 3H), 7.66 – 7.73 (m, 3H), 8.21 (s, 1H), 8.34 (t, *J* = 6.0 Hz, 2H), 8.60 (s, 1H), 8.68 (d, *J* = 2.3 Hz, 1H), 8.97 (s, 1H), 9.75 (s, 1H), 12.96 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 15.96, 16.80, 18.58, 18.85, 22.15, 25.67, 25.85, 26.09, 28.35, 28.85, 35.12, 37.01, 38.04, 41.69, 46.78, 53.47, 55.36, 57.76, 58.67, 67.56, 68.58, 69.59, 69.72, 69.75, 111.68, 112.28 (d, *J* = 23.3 Hz), 115.64, 117.50, 117.83 – 118.57 (m), 120.73, 121.82 – 122.26 (m), 122.97, 123.56, 126.81, 126.95, 127.00, 127.70, 127.86, 128.02, 128.36 – 129.05 (m), 130.95, 131.27, 131.31, 131.35, 131.53, 136.56, 138.66, 139.16, 142.17, 143.92, 147.85, 148.80, 151.37, 151.34 – 153.39 (m), 155.89, 155.93 (d, *J* = 240.5 Hz), 167.44, 168.08, 171.49, 172.05, 180.61; **LC-MS** (ESI) t_R = 11.78 min, 99% purity, *m/z* $[M + H]^+$ calcd for $C_{66}H_{77}F_2N_8O_{11}S_2$, 1259.51; found, 1259.5; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{66}H_{77}F_2N_8O_{11}S_2$, 1259.5116; found, 1259.5121.

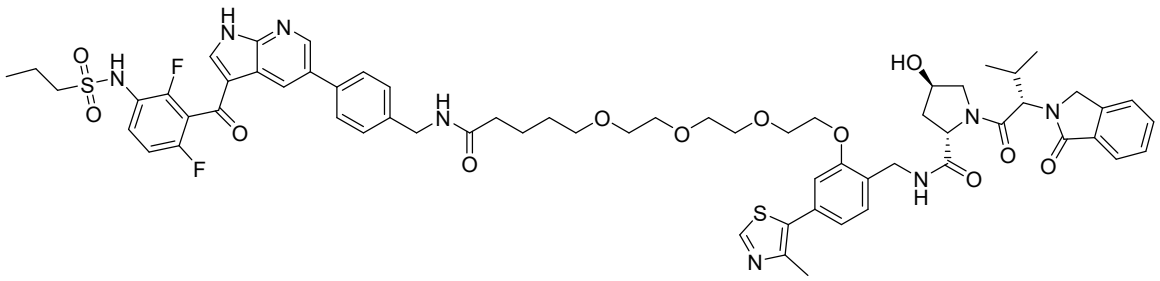
BRAF-targeting PROTAC 16b



This compound was synthesized by analogy with **16a** but using conjugate **14b** (82 mg, 0.1 mmol) and BRAF ligand **6** (58 mg, 0.1 mmol). The crude product was purified by column chromatography silica gel (gradient from CH_2Cl_2/MeOH 19:1 to 9:1), followed by preparative HPLC (H_2O/MeOH 25/75) to yield the title compound as a colourless solid.

Yield (77 mg, 64%); R_f = 0.22 (CH_2Cl_2/MeOH 15:1); mp 190 – 192 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.73 (d, *J* = 6.6 Hz, 3H), 0.93 – 0.99 (m, 6H), 1.70 – 1.78 (m, 2H), 1.89 – 1.96 (m, 1H), 2.01 – 2.07 (m, 1H), 2.28 – 2.36 (m, 1H), 2.46 (s, 3H), 3.10 – 3.15 (m, 2H), 3.45 (q, *J* = 5.9 Hz, 2H), 3.53 – 3.59 (m, 8H), 3.63 (dd, *J* = 3.7, 5.8 Hz, 2H), 3.67 – 3.71 (m, 1H), 3.75 – 3.82 (m, 3H), 4.18 (dd, *J* = 3.7, 5.7 Hz, 2H), 4.22 – 4.28 (m, 1H), 4.29 – 4.36 (m, 2H), 4.38 – 4.48 (m, 2H), 4.54 (d, *J* = 18.0 Hz, 1H), 4.71 (d, *J* = 10.8 Hz, 1H), 5.07 (d, *J* = 4.1 Hz, 1H), 7.00 (dd, *J* = 1.6, 7.7 Hz, 1H), 7.04 (d, *J* = 1.6 Hz, 1H), 7.25 – 7.31 (m, 1H), 7.34 (d, *J* = 7.8 Hz, 1H), 7.45 – 7.53 (m, 1H), 7.55 – 7.64 (m, 3H), 7.68 – 7.73 (m, 1H), 7.85 (d, *J* = 8.2 Hz, 2H), 7.99 (d, *J* = 8.4 Hz, 2H), 8.24 (s, 1H), 8.34 (t, *J* = 6.0 Hz, 1H), 8.59 (t, *J* = 5.6 Hz, 1H), 8.68 (s, 1H), 8.76 (d, *J* = 2.2 Hz, 1H), 8.96 (s, 1H), 9.75 (s, 1H), 13.02 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 15.97, 16.80, 18.58, 18.84, 28.35, 37.05, 38.06, 46.78, 53.48, 55.38, 57.76, 58.69, 67.90, 68.60, 68.90, 68.98, 69.62, 69.76, 69.83, 70.06, 112.17, 112.10 – 112.52 (m), 115.72, 117.48, 117.88 – 118.44 (m), 121.04, 121.72 – 122.14 (m), 122.97, 123.56, 126.87, 127.17, 127.68, 127.85, 128.03, 128.57 – 128.90 (m), 130.54, 130.96, 131.21, 131.35, 131.52, 133.27, 138.88, 140.67, 142.16, 144.10, 147.89, 149.10, 151.37, 150.96 – 153.70 (m), 155.85, 156.00 (d, *J* = 246.2 Hz), 165.86, 167.44, 168.09, 171.52, 180.64; **LC-MS** (ESI) t_R = 11.13 min, 99% purity, *m/z* $[M + H]^+$ calcd for $C_{61}H_{67}F_2N_8O_{12}S_2$, 1205.43; found, 1205.9; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{61}H_{67}F_2N_8O_{12}S_2$, 1205.4282; found, 1205.4289.

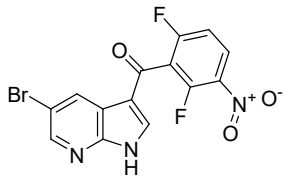
BRAF-targeting PROTAC 16c



This compound was synthesized by analogy with **16a** but using conjugate **14c** (84 mg, 0.1 mmol). The crude product was purified by column chromatography silica gel (gradient from CH_2Cl_2/MeOH 19:1 to 9:1), followed by preparative HPLC (H_2O/MeOH 25/75) to yield the title compound as a colourless solid.

Yield (52 mg, 42%); R_f = 0.42 (CH_2Cl_2/MeOH 12:1); mp 116 – 118 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.73 (d, *J* = 6.6 Hz, 3H), 0.94 – 0.98 (m, 6H), 1.46 – 1.52 (m, 2H), 1.54 – 1.60 (m, 2H), 1.70 – 1.78 (m, 2H), 1.89 – 1.95 (m, 1H), 2.01 – 2.07 (m, 1H), 2.17 (t, *J* = 7.4 Hz, 2H), 2.29 – 2.36 (m, 1H), 2.46 (s, 3H), 3.09 – 3.15 (m, 2H), 3.37 (t, *J* = 6.4 Hz, 2H), 3.44 – 3.47 (m, 2H), 3.50 – 3.57 (m, 4H), 3.60 – 3.64 (m, 2H), 3.66 – 3.81 (m, 4H), 4.18 (t, *J* = 4.7 Hz, 2H), 4.22 – 4.36 (m, 5H), 4.39 – 4.58 (m, 3H), 4.71 (d, *J* = 10.8 Hz, 1H), 5.07 (d, *J* = 4.0 Hz, 1H), 7.00 (dd, *J* = 1.6, 7.8 Hz, 1H), 7.05 (d, *J* = 1.6 Hz, 1H), 7.25 – 7.30 (m, 1H), 7.34 (d, *J* = 7.8 Hz, 1H), 7.38 (d, *J* = 7.9 Hz, 2H), 7.47 – 7.52 (m, 1H), 7.56 – 7.63 (m, 3H), 7.70 (dd, *J* = 7.8, 10.4 Hz, 3H), 8.21 (s, 1H), 8.31 – 8.36 (m, 2H), 8.60 (s, 1H), 8.68 (d, *J* = 2.2 Hz, 1H), 8.97 (s, 1H), 9.74 (s, 1H), 12.97 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.57, 15.98, 16.79, 18.58, 18.85, 22.09, 28.35, 28.79, 35.10, 37.04, 38.06, 41.70, 46.78, 53.48, 55.37, 57.76, 58.68, 67.90, 68.59, 68.97, 69.44, 69.80, 69.84, 69.99, 70.05, 112.18, 112.13 – 112.51 (m), 115.64, 117.49, 117.91 – 118.41 (m), 121.05, 121.95 (d, *J* = 14.4 Hz), 122.97, 123.57, 126.81, 127.00, 127.18, 127.69, 127.86, 128.03, 128.76, 130.95, 131.21, 131.32, 131.35, 131.53, 136.56, 138.66, 139.14, 142.17, 143.94, 147.89, 148.80, 151.39, 152.31 (d, *J* = 241.1 Hz), 155.86, 155.98 (d, *J* = 239.1 Hz), 167.45, 168.08, 171.52, 172.05, 180.59; **LC-MS** (ESI) t_R = 11.29 min, 99% purity, *m/z* $[M + H]^+$ calcd for $C_{64}H_{73}F_2N_8O_{12}S_2$, 1248.48; found, 1247.9; **HRMS** (ESI) *m/z* $[M + Na]^+$ calcd for $C_{64}H_{72}F_2N_8NaO_{12}S_2$, 1269.4571; found, 1269.4571.

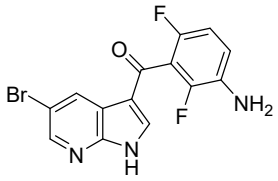
(5-Bromo-1*H*-pyrrolo[2,3-*b*]pyridin-3-yl)-(2,6-difluoro-3-nitrophenyl)methanone (18)



2,6-Difluoro-3-nitrobenzoic acid (**17**, 4.06 g, 20 mmol) was dissolved in dry CH_2Cl_2 (100 mL). Oxalyl chloride (2M in CH_2Cl_2, 30 mL, 60 mmol) and dry DMF (5 drops) were added and the mixture was stirred under argon atmosphere for 5 h. The solvent was removed under reduced pressure. 5-Bromo-7-azaindole (**2**, 5.92 g, 30 mmol) was suspended in dry CH_2Cl_2 (150 mL) and $AlCl_3$ (20.0 g, 150 mmol) was added in portions at 0 °C. Subsequently, the *in situ* generated acid chloride in dry CH_2Cl_2 (50 mL) was added dropwise, and the reaction mixture was stirred for 16 h under argon and was allowed to warm up to rt slowly. Subsequently, the mixture was poured onto a mixture of ice-cold brine and saturated $NaHCO_3$ solution. It was extracted with EtOAc (3 × 300 mL), washed with H_2O (200 mL) and brine (200 mL), dried over Na_2SO_4, filtered and evaporated. After purification by column chromatography (gradient of petroleum ether/EtOAc 1:1 to EtOAc), **18** was obtained as a pale brown solid.

Yield (3.74 g, 49%); R_f = 0.44 (petroleum ether/EtOAc 1:1); mp >250 °C; **^{1}H NMR** (500 MHz, DMSO-d_6) δ 7.45 – 7.62 (m, 1H), 8.43 (d, *J* = 7.6 Hz, 2H), 8.51 (d, *J* = 2.2 Hz, 1H), 8.64 (d, *J* = 2.3 Hz, 1H), 13.17 (br s, 1H); **^{13}C NMR** (126 MHz, DMSO-d_6) δ 113.45 (dd, *J* = 4.0, 23.9 Hz), 114.57, 114.83, 119.09, 119.32, 119.50, 129.21 (d, *J* = 11.4 Hz), 131.35, 134.50, 140.37, 145.63, 148.06, 150.02 – 164.42 (m), 178.99; **LC-MS** (ESI) t_R = 11.22 min, 97% purity, *m/z* $[M + H]^+$ calcd for $C_{14}H_7{}^{79}BrF_2N_3O_3$, 381.96; found, 381.9.

(3-Amino-2,6-difluorophenyl)-(5-bromo-1*H*-pyrrolo[2,3-*b*]pyridin-3-yl)methanone (19)

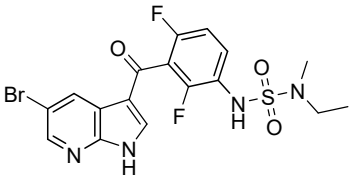


Iron (3.57 g, 64 mmol) was suspended in H_2O (80 mL) and AcOH (80 mL). Subsequently, a solution of nitro compound **18** (3.06 g, 8 mmol) in EtOH (80 mL) and AcOH (20 mL) was added slowly. The mixture was heated to 110 °C for 30 min, after which it was cooled to rt. The mixture was diluted with EtOAc (400 mL) and stirred with $NaHCO_3$ solution (400 mL; caution, CO_2 evolution!). The organic layer was separated and the aqueous phase was extracted again with EtOAc (2 × 400 mL). The combined organic layers were

washed with $NaHCO_3$ solution (400 mL), dried over Na_2SO_4, filtered, and concentrated *in vacuo* to give a light orange solid.

Yield (2.54 g, 90%); R_f = 0.37 (petroleum ether/EtOAc 1:2); mp 250 – 252 °C (dec.); **^{1}H NMR** (600 MHz, DMSO-d_6) δ 5.19 (s, 2H), 6.85 – 6.96 (m, 2H), 8.12 (s, 1H), 8.48 (d, *J* = 2.2 Hz, 1H), 8.54 (d, *J* = 2.4 Hz, 1H), 13.01 (br s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 111.40 – 111.73 (m), 114.23, 115.31, 116.73 – 117.08 (m), 117.18 – 117.64 (m), 119.24, 131.21, 133.59 (d, *J* = 12.9 Hz), 138.80, 145.22, 147.86, 145.25 – 150.13 (m), 182.34; **LC-MS** (ESI) t_R = 10.34 min, 96% purity, *m*/*z* [M + H]$^+$ calcd for $C_{14}H_9BrF_2N_3O$, 351.99; found, 351.8.

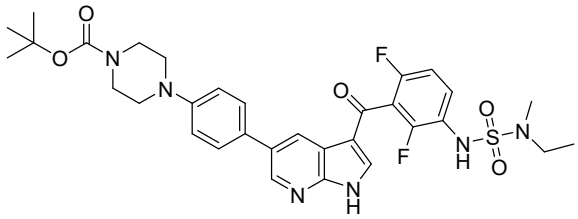
5-Bromo-3-[3-[[ethyl(methyl)sulfamoyl]amino]-2,6-difluorobenzoyl]-1*H*-pyrrolo[2,3-*b*]pyridine (20)



Compound **19** (1.06 g, 3 mmol) and *N*-ethyl-*N*-methylsulfamoyl chloride (1.42 g, 1.11 mL, 9 mmol) were dissolved in pyridine (30 mL) and heated to 65 °C for 16 h. The dark solution was evaporated *in vacuo* and the residue was diluted with half-saturated NH_4Cl solution (100 mL). It was extracted with 10% MeOH in EtOAc (2 × 100 mL), washed with brine (100 mL), dried over Na_2SO_4, filtered, and concentrated *in vacuo*. The crude product was purified by flash chromatography on spherical silica gel (50 g, 30 µm, gradient from 0% to 10% MeOH in $CHCl_3$) to yield the title compound as a light yellow solid.

Yield (0.51 g, 36%); R_f = 0.39 (10% MeOH in $CHCl_3$); mp >250 °C; **^{1}H NMR** (500 MHz, DMSO-d_6) δ 1.02 (t, *J* = 7.1 Hz, 3H), 2.72 (s, 3H), 3.11 (q, *J* = 7.1 Hz, 2H), 7.26 (td, *J* = 1.5, 8.9 Hz, 1H), 7.57 (td, *J* = 5.8, 9.0 Hz, 1H), 8.16 (s, 1H), 8.50 (d, *J* = 2.2 Hz, 1H), 8.55 (d, *J* = 2.3 Hz, 1H), 9.66 (s, 1H), 13.08 (s, 1H); **^{13}C NMR** (126 MHz, DMSO-d_6) δ 12.95, 34.12, 45.00, 111.95 – 112.82 (m), 114.39, 115.06, 117.38 – 118.59 (m), 119.15, 122.69 (d, *J* = 13.7 Hz), 128.37 (d, *J* = 9.7 Hz), 131.20, 139.13, 145.44, 147.91, 150.34 – 157.42 (m), 180.79; **LC-MS** (ESI) t_R = 6.50 min, 99% purity, *m*/*z* [M + H]$^+$ calcd for $C_{17}H_{16}{}^{79}BrF_2N_4O_3S$, 473.01; found, 473.2; **HRMS** (ESI) *m*/*z* [M + H]$^+$ calcd for $C_{15}H_{19}N_2O_4$, 473.0089; found, 473.0083.

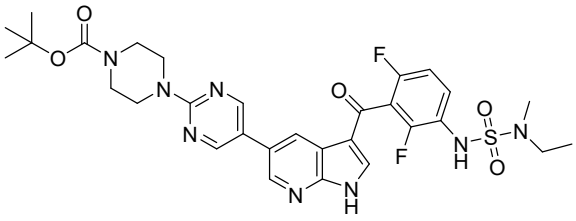
***tert*-Butyl 4-[4-[3-[3-[[ethyl(methyl)sulfamoyl]amino]-2,6-difluorobenzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl]phenyl]piperazine-1-carboxylate (21)**



Compound **20** (236 mg, 0.5 mmol) was dissolved in dry dioxane (20 mL) and the solution was purged with argon. 4-(4-Boc-piperazino)phenylboronic acid pinacol ester (233 mg, 0.6 mmol), K_2CO_3 (207 mg, 1.5 mmol), tricyclohexylphosphine (14 mg, 0.05 mmol), and bis(dibenzylidenacetone)palladium(0) (14 mg, 0.025 mmol) were added. The mixture was heated at 90 °C 3 h, after which a second portion of $Pd(dba)_2$ (14 mg) was added. After stirring for another 3 h at 90 °C, it was cooled, diluted with EtOAc (100 mL), and washed with half-saturated brine (50 mL). The organic layer was dried over Na_2SO_4, filtered, and concentrated *in vacuo.* The crude product was purified by flash chromatography on spherical silica gel (50 g, 30 µm, gradient from 10% to 100% EtOAc in cyclohexane) to yield the title compound as a light yellow solid.

Yield (154 mg, 47%); R_f = 0.26 (80% EtOAc in cyclohexane); mp 216 – 220 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 1.00 (t, *J* = 7.1 Hz, 3H), 1.42 (s, 9H), 2.71 (s, 3H), 3.10 (q, *J* = 7.1 Hz, 2H), 3.18 (dd, *J* = 4.0, 6.3 Hz, 4H), 3.48 (t, *J* = 5.2 Hz, 4H), 7.06 – 7.10 (m, 2H), 7.26 (td, *J* = 1.5, 8.8 Hz, 1H), 7.53 – 7.63 (m, 3H), 8.07 (s, 1H), 8.52 (s, 1H), 8.65 (d, *J* = 2.3 Hz, 1H), 9.67 (s, 1H), 12.89 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.98, 28.24, 34.13, 45.03, 48.22, 79.17, 111.89 – 112.97 (m), 115.73, 116.46, 117.71, 118.32 (t, *J* = 23.2 Hz), 122.66 (d, *J* = 13.3 Hz), 126.16, 127.80, 128.22 (d, *J* = 9.8 Hz), 128.86, 131.63, 138.29, 143.76, 148.54, 150.49, 152.12 (dd, *J* = 8.7, 249.8 Hz), 154.05, 155.77 (dd, *J* = 6.9, 245.9 Hz), 180.75; **LC-MS** (ESI) t_R = 7.85 min, 96% purity, *m/z* $[M + H]^+$ calcd for $C_{32}H_{37}F_2N_6O_5S$, 655.25; found, 655.4; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{32}H_{37}F_2N_6O_5S$, 665.2509; found, 665.2493.

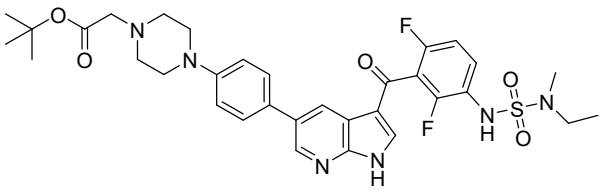
***tert*-Butyl 4-[5-[3-[3-[[ethyl(methyl)sulfamoyl]amino]-2,6-difluorobenzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl]pyrimidin-2-yl]piperazine-1-carboxylate (22)**



This compound was synthesized by analogy with **21** but using 2-(4-Boc-piperazino)pyrimidine-5-boronic acid pinacol ester (234 mg, 0.6 mmol). The crude product was purified by flash chromatography on spherical silica gel (50 g, 30 µm, gradient from 50% to 100% EtOAc in cyclohexane) to yield the title compound as a light yellow solid.

Yield (171 mg, 52%); R_f = 0.38 (80% EtOAc in cyclohexane); mp >250 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 1.01 (t, *J* = 7.1 Hz, 3H), 1.43 (s, 9H), 2.72 (s, 3H), 3.10 (q, *J* = 7.1 Hz, 2H), 3.43 (t, *J* = 5.3 Hz, 4H), 3.77 – 3.82 (m, 4H), 7.23 – 7.29 (m, 1H), 7.57 (td, *J* = 5.9, 9.0 Hz, 1H), 8.11 (s, 1H), 8.54 (s, 1H), 8.65 (d, *J* = 2.2 Hz, 1H), 8.77 (s, 2H), 9.67 (s, 1H), 12.97 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.97, 28.24, 34.12, 43.47, 45.02, 79.25, 112.37 (d, *J* = 22.9 Hz), 115.74, 117.67, 117.88 – 118.89 (m), 120.82, 122.30 – 123.15 (m), 126.26, 126.64, 128.29 (d, *J* = 9.6 Hz), 138.52, 143.41, 148.87, 152.14 (dd, *J* = 8.7, 249.7 Hz), 154.11, 155.77 (dd, *J* = 6.8, 246.2 Hz), 156.36, 160.64, 180.73; **LC-MS** (ESI) t_R = 7.69 min, 99% purity, *m*/*z* [M + H]$^+$ calcd for $C_{30}H_{35}F_2N_8O_5S$, 657.24; found, 655.4; **HRMS** (ESI) *m*/*z* [M + H]$^+$ calcd for $C_{30}H_{35}F_2N_8O_5S$, 657.2414; found, 657.2402.

***tert*-Butyl 2-[4-[4-[3-[3-[[ethyl(methyl)sulfamoyl]amino]-2,6-difluorobenzoyl]-1*H*-pyrrolo[2,3-*b*]pyridin-5-yl]phenyl]piperazin-1-yl]acetate (23)**

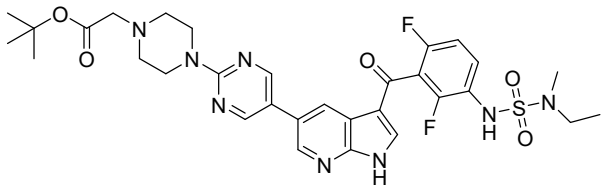


Compound **21** (0.15 g, 0.23 mmol) was treated with 4N HCl in dioxane (5 mL) for 3 h at rt. The orange solid was filtered, washed with dry dioxane (2 × 5 mL), and the salt was dried *in vacuo*. The material was dissolved in dry DMF (10 mL), DIPEA (0.12 mL, 0.69 mmol) and *tert*-butyl bromoacetate (37 µL, 0.25 mmol), and the mixture was stirred at rt for 3 h. It was diluted with CH_2Cl_2 (30 mL), washed with

saturated NH_4Cl solution, and extracted again with CH_2Cl_2 (30 mL). The combined organic layers were washed with 5% LiCl solution and brine (each 50 mL), dried over Na_2SO_4, filtered, and evaporated. The crude product was purified by flash chromatography on spherical silica gel (25 g, 15 µm, gradient from 0% to 7% MeOH in CH_2Cl_2) to yield the title compound as an orange solid.

Yield (94 mg, 61%); R_f = 0.45 (7% MeOH in CH_2Cl_2); mp 202 – 204 °C; **^{1}H NMR** (500 MHz, DMSO-d_6) δ 1.01 (t, *J* = 7.1 Hz, 3H), 1.42 (s, 9H), 2.66 (t, *J* = 5.0 Hz, 4H), 2.72 (s, 3H), 3.10 (q, *J* = 7.1 Hz, 2H), 3.17 (s, 2H), 3.21 (t, *J* = 5.0 Hz, 4H), 7.03 – 7.09 (m, 2H), 7.26 (td, *J* = 1.5, 8.7 Hz, 1H), 7.53 – 7.61 (m, 3H), 8.06 (s, 1H), 8.51 (s, 1H), 8.64 (d, *J* = 2.2 Hz, 1H), 9.66 (s, 1H), 12.87 (s, 1H); **^{13}C NMR** (126 MHz, DMSO-d_6) δ 12.95, 27.96, 34.11, 45.00, 48.09, 51.94, 59.39, 80.35, 112.31 (d, *J* = 22.0 Hz), 115.70, 115.93, 117.69, 118.32 (t, *J* = 23.5 Hz), 122.68 (d, *J* = 14.2 Hz), 126.03, 127.71, 127.80 – 128.44 (m), 128.30, 131.69, 138.17, 143.70, 148.47, 150.57, 152.09 (dd, *J* = 8.2, 249.8 Hz), 154.34 – 157.52 (m), 169.37, 180.72.; **LC-MS** (ESI) t_R = 7.86 min, 98% purity, *m/z* $[M + H]^+$ calcd for $C_{33}H_{39}F_2N_6O_5S$, 669.27; found, 669.5; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{33}H_{39}F_2N_6O_5S$, 669.2665; found, 669.2651.

tert-Butyl 2-[4-[5-[3-[3-[[ethyl(methyl)sulfamoyl]amino]-2,6-difluoro-benzoyl]-1_H_-pyrrolo[2,3-_b_]pyridin-5-yl]pyrimidin-2-yl]piperazin-1-yl]acetate (24)

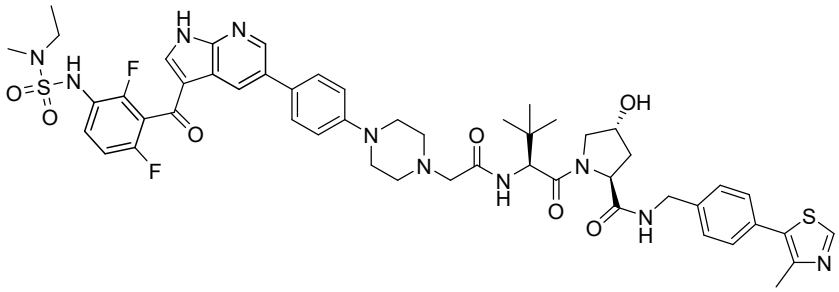


This compound was synthesized by analogy with **23** but using precursor 22 (0.15 g, 0.23 mmol). The crude product was purified by flash chromatography on spherical silica gel (25 g, 15 µm, gradient from 0% to 7% MeOH in CH_2Cl_2) to yield the title compound as a colourless solid.

Yield (79 mg, 51%); R_f = 0.29 (7% MeOH in CH_2Cl_2); mp 238 – 240 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ) 1.01 (t, *J* = 7.1 Hz, 3H), 1.41 (s, 9H), 2.57 – 2.62 (m, 4H), 2.72 (s, 3H), 3.10 (q, *J* = 7.1 Hz, 2H), 3.17 (s, 2H), 3.78 – 3.82 (m, 4H), 7.23 – 7.29 (m, 1H), 7.53 – 7.60 (m, 1H), 8.11 (s, 1H), 8.53 (s, 1H), 8.65 (d, *J* = 2.2 Hz, 1H), 8.74 (s, 2H), 9.67 (s, 1H), 12.96 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.98, 27.98, 34.13, 43.73, 45.03, 51.79, 59.33, 80.41, 111.83 – 112.78 (m), 115.73, 117.68, 118.02 – 118.57 (m), 120.52, 122.65 (d, *J* = 13.2 Hz), 126.18, 126.74, 128.28 (d, *J* = 9.9 Hz), 138.48, 143.39, 148.84, 152.14 (dd, *J* = 8.7, 249.7 Hz), 154.05 – 157.72 (m), 156.32, 160.72, 169.40, 180.73; **LC-MS** (ESI) t_R = 7.41 min, 99% purity, *m/z* $[M + H]^+$ calcd for

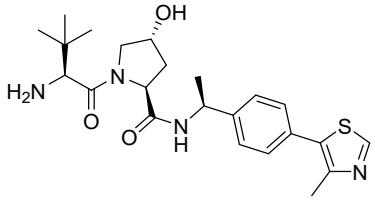
$C_{31}H_{37}F_2N_8O_5S$, 671.26; found, 671.4; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{31}H_{37}F_2N_8O_5S$, 671.2570; found, 671.2555.

Paradox-breaker PROTAC 25



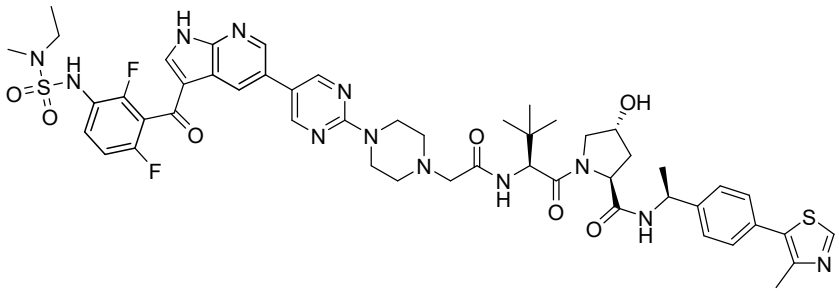
Yield (63 mg, 61%); R_f = 0.44 (10% MeOH in CH_2Cl_2); mp 212 – 214 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.96 (s, 9H), 1.01 (t, *J* = 7.1 Hz, 3H), 1.86 – 1.94 (m, 1H), 2.02 – 2.09 (m, 1H), 2.39 (s, 3H), 2.62 – 2.70 (m, 4H), 2.72 (s, 3H), 3.01 – 3.16 (m, 4H), 3.21 – 3.29 (m, 4H), 3.62 (d, *J* = 10.6 Hz, 1H), 3.67 (dd, *J* = 4.0, 10.7 Hz, 1H), 4.25 (dd, *J* = 5.7, 15.7 Hz, 1H), 4.34 – 4.48 (m, 3H), 4.53 (d, *J* = 9.6 Hz, 1H), 5.13 (d, *J* = 3.5 Hz, 1H), 7.05 – 7.11 (m, 2H), 7.23 – 7.29 (m, 1H), 7.34 – 7.43 (m, 4H), 7.53 – 7.62 (m, 3H), 7.83 (d, *J* = 9.6 Hz, 1H), 8.07 (s, 1H), 8.53 (s, 1H), 8.57 (t, *J* = 6.1 Hz, 1H), 8.64 (d, *J* = 2.3 Hz, 1H), 8.90 (s, 1H), 9.67 (s, 1H), 12.89 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.97, 16.04, 26.41, 34.12, 35.93, 38.02, 41.82, 45.02, 48.33, 52.91, 56.02, 56.69, 58.95, 60.76, 69.06, 112.34 (d, *J* = 20.1 Hz), 115.72, 115.99, 117.71, 118.32 (t, *J* = 23.5 Hz), 122.68 (d, *J* = 12.5 Hz), 126.10, 127.68, 127.73, 128.49, 128.06 – 129.26 (m), 128.79, 129.85, 131.28, 131.66, 138.27, 139.58, 143.73, 147.85, 148.51, 150.45, 151.45, 152.10 (dd, *J* = 8.0, 249.7 Hz), 155.74 (dd, *J* = 6.7, 246.3 Hz), 168.65, 169.46, 171.92, 180.75; **LC-MS** (ESI) t_R = 6.65 min, 98% purity, *m/z* $[M + H]^+$ calcd for $C_{51}H_{59}F_2N_{10}O_7S_2$, 1025.40; found, 1025.7; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{51}H_{59}F_2N_{10}O_7S_2$, 1025.3972; found, 1025.3958.

(2*S*,4*R*)-1-[(2*S*)-2-amino-3,3-dimethyl-butanoyl]-4-hydroxy-*N*-[(1*S*)-1-[4-(4-methylthiazol-5-yl)phenyl]ethyl]pyrrolidine-2-carboxamide (27)



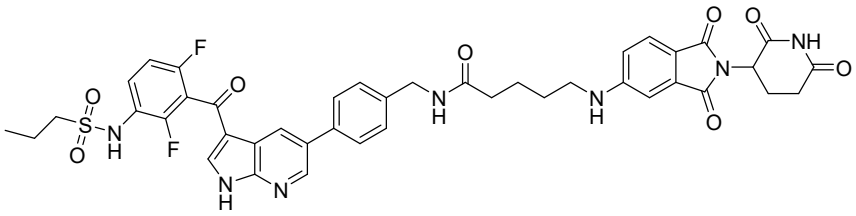
This compound was synthesized as we described previously.[13]

Paradox-breaker PROTAC 26



Yield (69 mg, 66%); R_f = 0.44 (10% MeOH in CH_2Cl_2); mp 208 – 210 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.96 (s, 9H), 1.01 (t, *J* = 7.1 Hz, 3H), 1.37 (d, *J* = 7.0 Hz, 3H), 1.74 – 1.81 (m, 1H), 2.02 – 2.09 (m, 1H), 2.44 (s, 3H), 2.58 (h, *J* = 6.8 Hz, 4H), 2.72 (s, 3H), 3.01 – 3.15 (m, 4H), 3.56 – 3.64 (m, 2H), 3.80 – 3.89 (m, 4H), 4.26 – 4.31 (m, 1H), 4.46 (t, *J* = 8.2 Hz, 1H), 4.52 (d, *J* = 9.6 Hz, 1H), 4.90 (p, *J* = 7.1 Hz, 1H), 5.11 (d, *J* = 3.5 Hz, 1H), 7.23 – 7.29 (m, 1H), 7.32 – 7.39 (m, 2H), 7.40 – 7.45 (m, 2H), 7.53 – 7.60 (m, 1H), 7.80 (d, *J* = 9.6 Hz, 1H), 8.11 (s, 1H), 8.42 (d, *J* = 7.7 Hz, 1H), 8.54 (s, 1H), 8.64 – 8.71 (m, 1H), 8.76 (s, 2H), 8.96 (s, 1H), 9.67 (s, 1H), 12.96 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.97, 16.14, 22.64, 26.48, 34.13, 35.88, 37.91, 43.88, 45.03, 47.94, 52.76, 56.07, 56.67, 58.70, 60.82, 68.94, 112.37 (d, *J* = 23.9 Hz), 115.75, 117.68, 118.26 (t, *J* = 23.2 Hz), 120.72, 122.67 (d, *J* = 11.3 Hz), 126.24, 126.47, 126.69, 128.28 (d, *J* = 10.1 Hz), 128.99, 129.04, 129.84, 131.27, 138.51, 143.42, 144.93, 147.91, 148.86, 151.60, 152.14 (dd, *J* = 8.1, 249.7 Hz), 155.77 (dd, *J* = 6.9, 246.1 Hz), 156.34, 160.71, 168.68, 169.38, 170.65, 180.74; **LC-MS** (ESI) t_R = 6.73 min, 99% purity, *m/z* $[M + H]^+$ calcd for $C_{50}H_{59}F_2N_{12}O_7S_2$, 1041.40; found, 1041.7; **HRMS** (ESI) *m/z* $[M + H]^+$ calcd for $C_{50}H_{59}F_2N_{12}O_7S_2$, 1041.4034; found, 1041.4019.

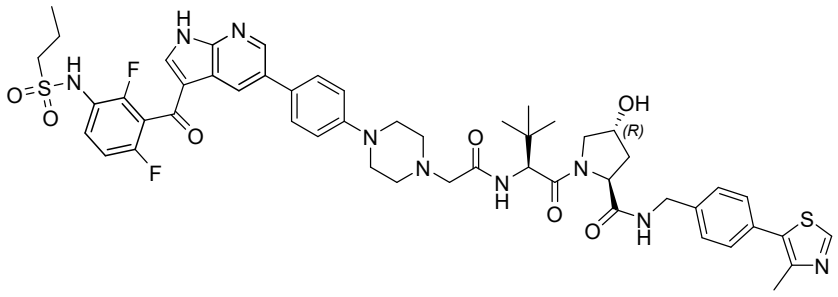
BRAF^{V600E} PROTAC "cmpd 12" [12]



This compound was synthesized as described previously.[12]

Yield (63%); R_f = 0.25 (5% MeOH/CH_2Cl_2); mp 174 – 176 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.95 (t, *J* = 7.4 Hz, 3H), 1.55 – 1.62 (m, 2H), 1.62 – 1.69 (m, 2H), 1.69 – 1.78 (m, 2H), 1.93 – 2.01 (m, 1H), 2.22 (t, *J* = 7.2 Hz, 2H), 2.44 – 2.59 (m, 2H), 2.81 – 2.90 (m, 1H), 3.08 – 3.14 (m, 2H), 3.18 (q, *J* = 6.5 Hz, 2H), 4.33 (d, *J* = 5.9 Hz, 2H), 5.01 (dd, *J* = 5.4, 12.9 Hz, 1H), 6.85 (dd, *J* = 2.1, 8.4 Hz, 1H), 6.95 (d, *J* = 2.2 Hz, 1H), 7.11 (t, *J* = 5.4 Hz, 1H), 7.24 – 7.30 (m, 1H), 7.38 (d, *J* = 7.9 Hz, 2H), 7.52 – 7.61 (m, 2H), 7.68 (d, *J* = 7.9 Hz, 2H), 8.21 (s, 1H), 8.37 (t, *J* = 6.0 Hz, 1H), 8.59 (s, 1H), 8.67 (d, *J* = 2.2 Hz, 1H), 9.74 (s, 1H), 11.02 (s, 1H), 12.96 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.75, 16.98, 22.38, 23.05, 28.03, 31.14, 35.16, 41.90, 42.37, 48.78, 53.66, 112.48 (d, *J* = 23.2 Hz), 115.24, 115.82, 116.00, 117.67, 118.05 – 119.00 (m), 122.18 (d, *J* = 12.8 Hz), 125.22, 126.99, 127.20, 128.22, 128.89 (d, *J* = 9.7 Hz), 131.48, 134.39, 136.76, 138.87, 139.29, 144.11, 148.98, 152.49 (dd, *J* = 8.7, 249.7 Hz), 154.62, 156.14 (dd, *J* = 6.9, 246.7 Hz), 167.29, 167.86, 170.30, 172.15, 172.93, 180.79; **LC-MS** (ESI) t_R = 6.02 min, 98% purity, *m*/*z* [M + H]$^+$ calcd for $C_{42}H_{40}F_2N_7O_8S$, 840.26; found, 840.5.

BRAF^{V600E} PROTAC SJF-0628 [16]

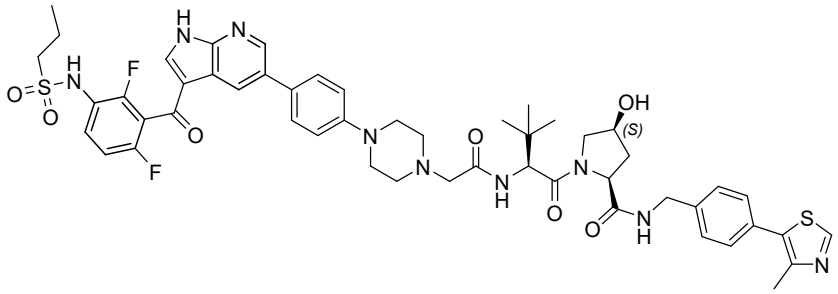


This compound was synthesized as described previously.[16]

Yield (64%); R_f = 0.31 (7% MeOH/CH_2Cl_2); mp 164 – 166 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.90 – 1.00 (m, 12H), 1.73 (h, *J* = 7.5 Hz, 2H), 1.86 – 1.94 (m, 1H), 2.02 – 2.09 (m, 1H), 2.39 (s, 3H), 2.61 – 2.72 (m, 4H), 3.01 – 3.16 (m, 4H), 3.21 – 3.29 (m, 4H), 3.60 – 3.71 (m, 2H), 4.25 (dd, *J* = 5.7, 15.8 Hz, 1H), 4.35 – 4.48 (m, 3H), 4.53 (d, *J* = 9.6 Hz, 1H), 5.13 (d, *J* = 3.5 Hz, 1H), 7.08 (d, *J* = 8.8 Hz, 2H), 7.26 (t, *J* = 8.6 Hz, 1H), 7.34 –

7.47 (m, 4H), 7.52 – 7.62 (m, 3H), 7.83 (d, *J* = 9.6 Hz, 1H), 8.16 (s, 1H), 8.54 (s, 1H), 8.57 (t, *J* = 6.1 Hz, 1H), 8.64 (d, *J* = 2.3 Hz, 1H), 8.90 (s, 1H), 9.74 (s, 1H), 12.89 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.77, 16.04, 17.00, 26.42, 35.94, 38.02, 41.83, 48.34, 52.91, 53.65, 56.03, 56.70, 58.96, 60.77, 69.06, 112.44 (d, *J* = 23.5 Hz), 115.77, 116.00, 117.74, 118.41 (t, *J* = 23.6 Hz), 121.85 – 122.84 (m), 126.12, 127.68, 127.73, 128.50, 128.19 – 129.16 (m), 128.80, 129.85, 131.28, 131.64, 138.57, 139.59, 143.71, 147.86, 148.55, 150.45, 151.45, 152.46 (dd, *J* = 8.2, 249.4 Hz), 154.76 – 157.38 (m), 168.67, 169.46, 171.93, 180.76; **LC-MS** (ESI) t_R = 6.81 min, 95% purity, *m*/*z* [M + H]$^+$ calcd for $C_{51}H_{58}F_2N_9O_7S_2$, 1010.39; found, 1010.7.

BRAF^{V600E} PROTAC SJF-0661 (negative control) [16]



This compound was synthesized as described previously.[16]

Yield (65%); R_f = 0.26 (7% MeOH/CH_2Cl_2); mp 204 – 210 °C; **^{1}H NMR** (600 MHz, DMSO-d_6) δ 0.95 (t, *J* = 7.4 Hz, 3H), 0.98 (s, 9H), 1.69 – 1.79 (m, 2H), 1.91 – 1.99 (m, 1H), 2.02 – 2.10 (m, 1H), 2.38 (s, 3H), 2.56 – 2.67 (m, 4H), 2.86 – 3.23 (m, 8H), 3.59 (dd, *J* = 3.3, 10.1 Hz, 1H), 3.75 (dd, *J* = 4.8, 10.4 Hz, 1H), 4.28 – 4.45 (m, 4H), 4.50 (d, *J* = 9.0 Hz, 1H), 5.12 (d, *J* = 3.8 Hz, 1H), 6.93 – 6.98 (m, 2H), 7.24 – 7.30 (m, 1H), 7.35 – 7.43 (m, 4H), 7.52 (d, *J* = 8.4 Hz, 2H), 7.54 – 7.62 (m, 1H), 7.72 (d, *J* = 8.8 Hz, 1H), 8.16 (s, 1H), 8.32 (t, *J* = 6.0 Hz, 1H), 8.52 (s, 1H), 8.61 (d, *J* = 2.2 Hz, 1H), 8.91 (s, 1H), 9.72 (s, 1H), 12.89 (s, 1H); **^{13}C NMR** (151 MHz, DMSO-d_6) δ 12.75, 16.08, 16.97, 26.53, 34.94, 38.06, 41.76, 48.28, 52.72, 53.66, 55.68, 56.30, 58.87, 60.77, 68.71, 112.48 (d, *J* = 23.0 Hz), 115.76, 115.87, 117.72, 118.42 (t, *J* = 23.6 Hz), 121.70 – 122.42 (m), 127.62, 127.65, 127.95 – 129.27 (m), 128.93, 129.97, 131.24, 131.63, 138.57, 139.51, 143.66, 147.91, 148.53, 150.41, 151.46, 152.50 (dd, *J* = 8.1, 249.3 Hz), 156.18 (dd, *J* = 6.7, 246.5 Hz), 169.41, 169.45, 171.66, 180.71; **LC-MS** (ESI) t_R = 6.46 min, 97% purity, *m*/*z* [M + H]$^+$ calcd for $C_{51}H_{58}F_2N_9O_7S_2$, 1010.39; found, 1010.6.

K. Crystallographic data and structure refinement

Crystal Habitus	clear colourless plank
Device Type	Bruker X8-KappaApexII
Empirical formula	$C_{17}H_{14}N_3O_3F_2SBr$
Formula weight	458.28
Temperature/K	100
Crystal system	Triclinic
Space group	P-1
a/Å	6.4327(4)
b/Å	7.6898(6)
c/Å	18.8782(14)
α/°	90.654(4)
β/°	91.728(4)
γ/°	103.495(4)
Volume/Å^3	907.47(11)
Z	2
ρ_{calc}g/cm^3	1.677
μ/mm^{-1}	2.423
F(000)	460.0
Crystal size/mm^3	0.22 × 0.08 × 0.02
Absorption correction	empirical
Tmin; Tmax	0.6154; 0.7460
Radiation	MoKα (λ = 0.71073)
2Θ range for data collection/°	5.448 to 55.996°
Completeness to theta	0.998
Index ranges	-8 ≤ h ≤ 8, -10 ≤ k ≤ 10, -24 ≤ l ≤ 24
Reflections collected	33623
Independent reflections	4393 [R_{int} = 0.0848, R_{sigma} = 0.0537]
Data/restraints/parameters	4393/0/245
Goodness-of-fit on F^2	1.009
Final R indexes [I>=2σ (I)]	R_1 = 0.0344, wR_2 = 0.0745
Final R indexes [all data]	R_1 = 0.0609, wR_2 = 0.0841
Largest diff. peak/hole / e Å^{-3}	0.74/-0.79

Bond lengths

Atom	Atom	Length/Å	Atom	Atom	Length/Å
Br	C4	1.891(3)	C1	C8	1.439(4)
S	O2	1.431(2)	C2	C3	1.394(3)
S	O3	1.4414(19)	C2	C6	1.413(4)
S	N3	1.640(2)	C3	C4	1.378(4)
S	C15	1.755(3)	C4	C5	1.396(4)
F1	C10	1.350(3)	C8	C9	1.517(4)
F2	C14	1.357(3)	C9	C10	1.386(4)
O1	C8	1.218(3)	C9	C14	1.379(4)
N1	C6	1.375(3)	C10	C11	1.383(4)
N1	C7	1.341(4)	C11	C12	1.377(4)
N2	C5	1.335(3)	C12	C13	1.387(4)
N2	C6	1.329(3)	C13	C14	1.377(4)
N3	C11	1.430(3)	C15	C16	1.514(4)
C1	C2	1.438(4)	C16	C17	1.518(5)
C1	C7	1.383(4)			

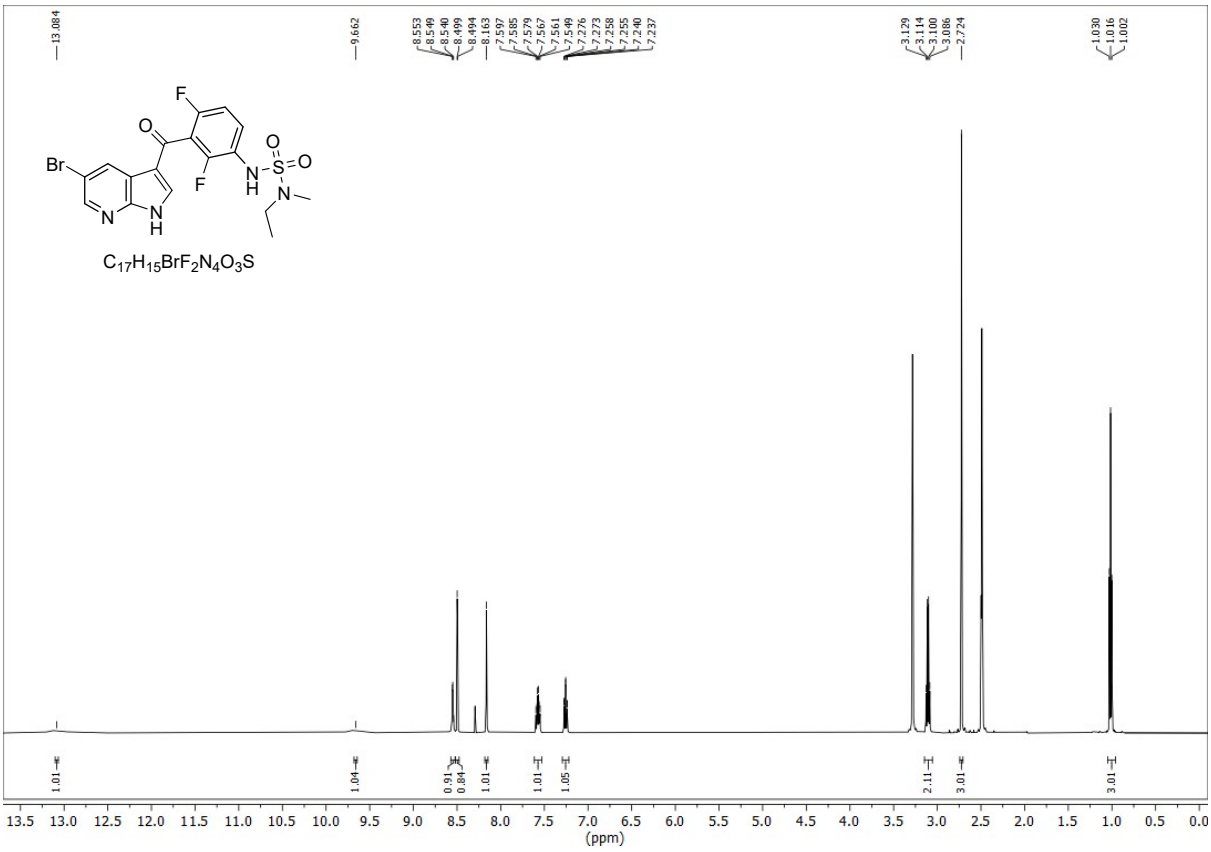
Bond angles

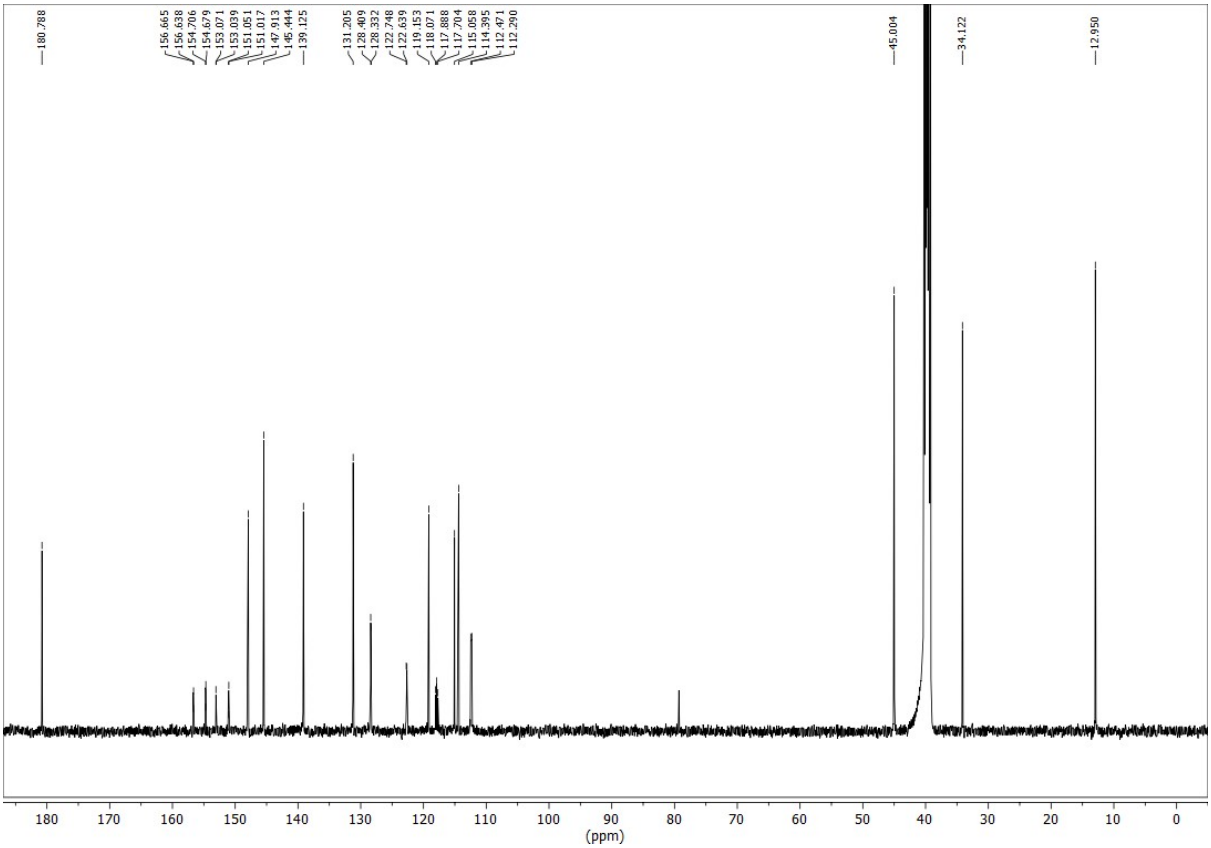
Atom	Atom	Atom	Angle/˚	Atom	Atom	Atom	Angle/˚
O2	S	O3	118.95(13)	N2	C6	C2	126.3(2)
O2	S	N3	109.59(12)	N1	C7	C1	110.9(2)
O2	S	C15	108.86(13)	O1	C8	C1	124.1(3)
O3	S	N3	104.66(12)	O1	C8	C9	118.7(2)
O3	S	C15	108.46(13)	C1	C8	C9	117.1(2)
N3	S	C15	105.50(13)	C10	C9	C8	122.9(3)
C7	N1	C6	108.7(2)	C14	C9	C8	121.3(2)
C6	N2	C5	115.2(2)	C14	C9	C10	115.8(2)
C11	N3	S	119.38(18)	F1	C10	C9	118.2(2)
C2	C1	C8	126.9(2)	F1	C10	C11	118.7(2)
C7	C1	C2	106.1(2)	C11	C10	C9	123.0(3)
C7	C1	C8	127.1(2)	C10	C11	N3	120.9(3)

C3	C2	C1	136.8(2)	C12	C11	N3	120.6(2)
C3	C2	C6	117.4(2)	C12	C11	C10	118.5(3)
C6	C2	C1	105.8(2)	C11	C12	C13	120.9(3)
C4	C3	C2	116.5(2)	C14	C13	C12	118.0(3)
C3	C4	Br	119.90(19)	F2	C14	C9	117.6(2)
C3	C4	C5	121.8(2)	F2	C14	C13	118.6(3)
C5	C4	Br	118.3(2)	C13	C14	C9	123.8(3)
N2	C5	C4	122.8(3)	C16	C15	S	114.7(2)
N1	C6	C2	108.5(2)	C15	C16	C17	110.3(3)
N2	C6	N1	125.2(2)				

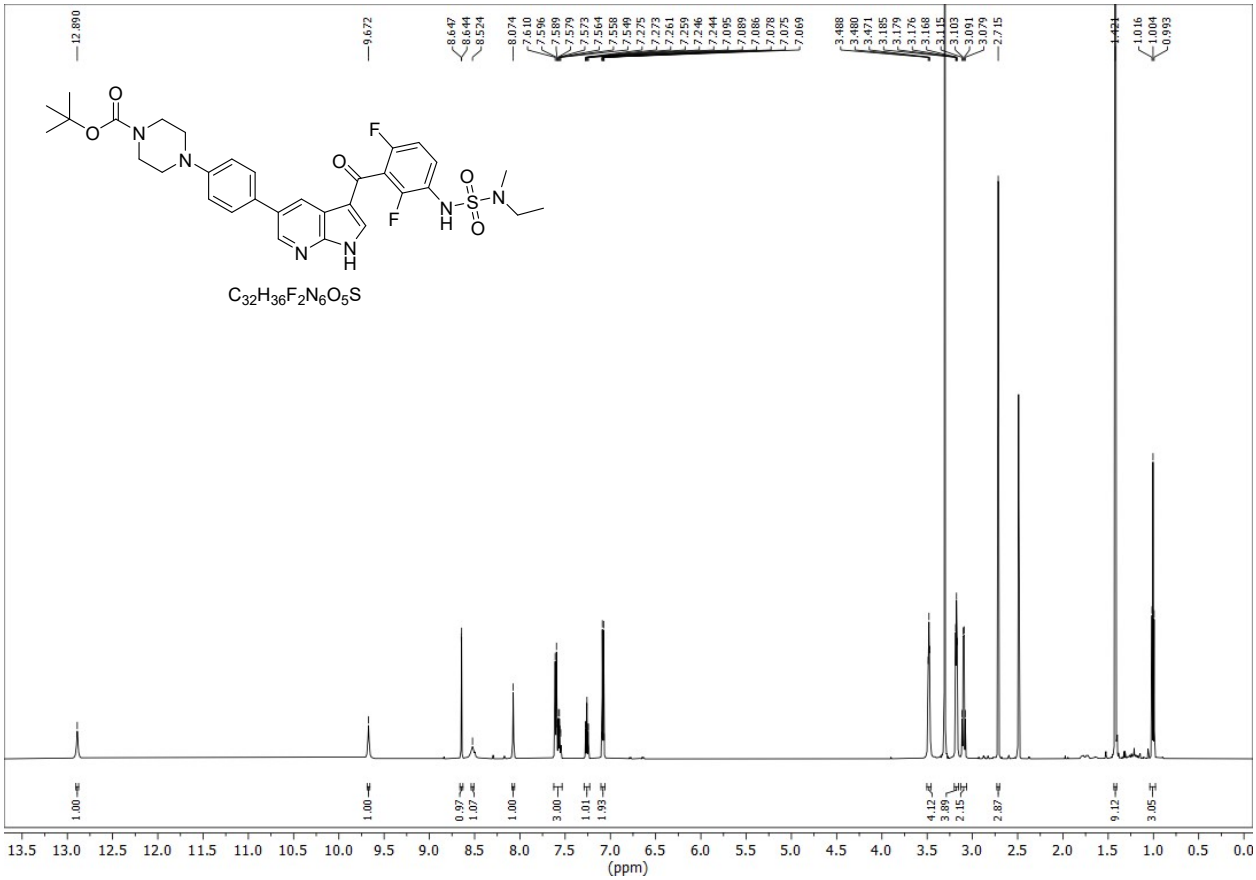
L. Selected NMR and LC/MS traces

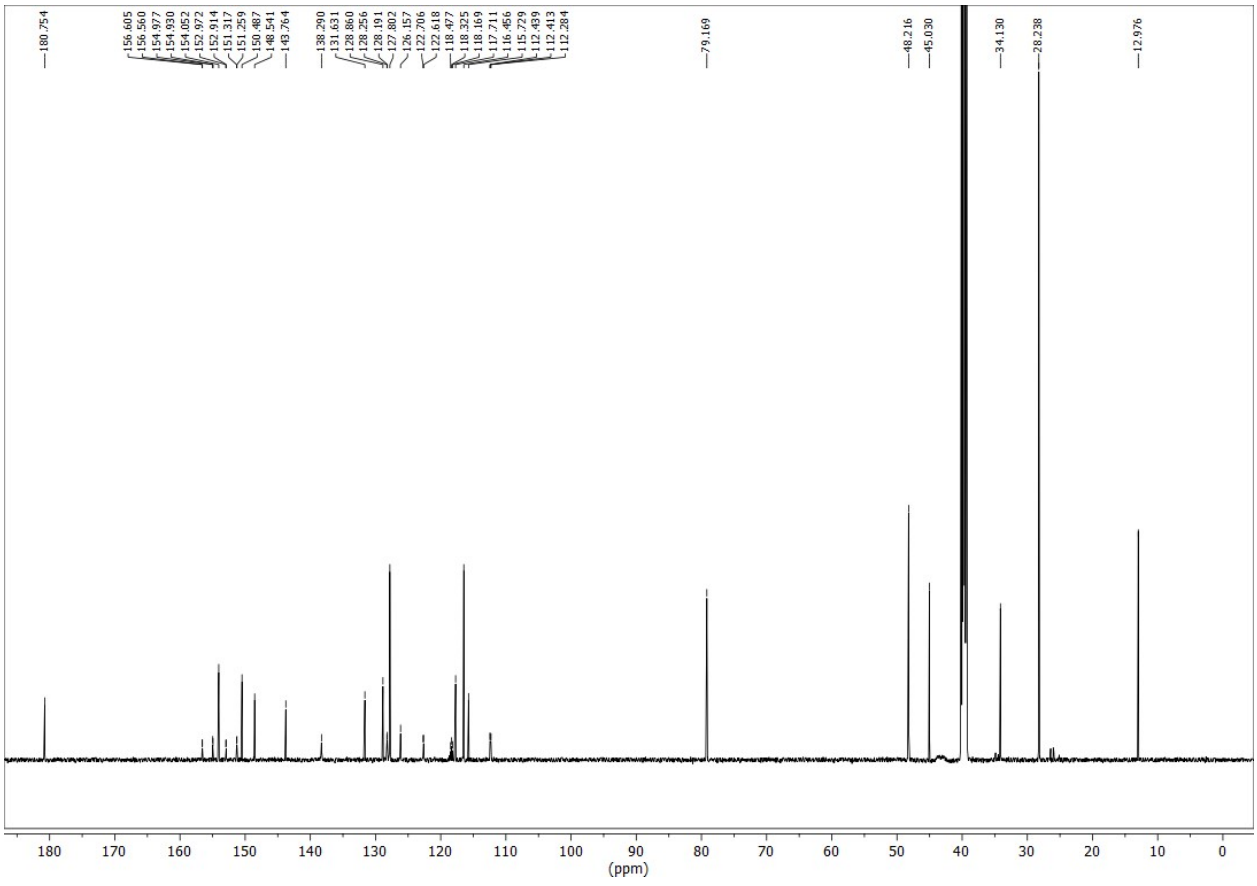
^{1}H and ^{13}C NMR spectrum of compound **20**.



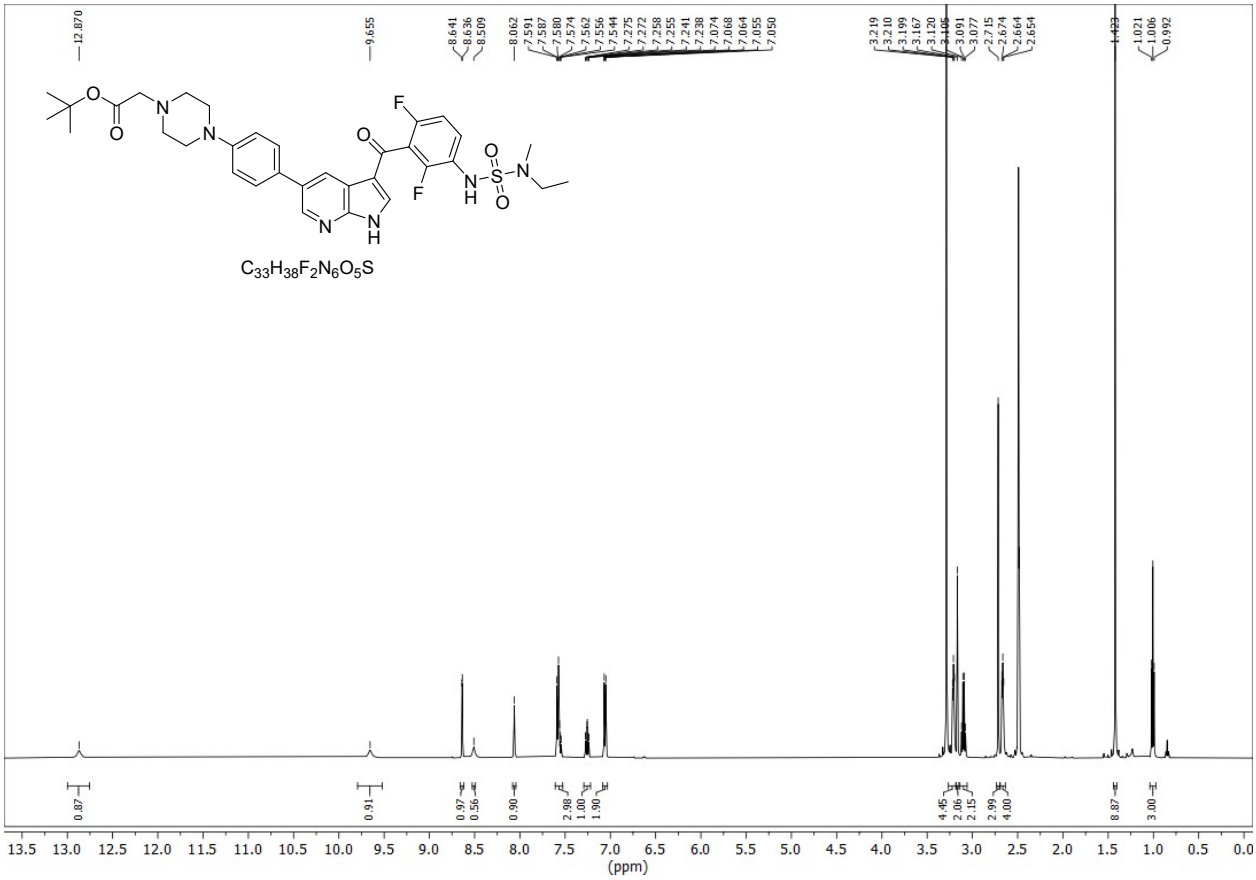


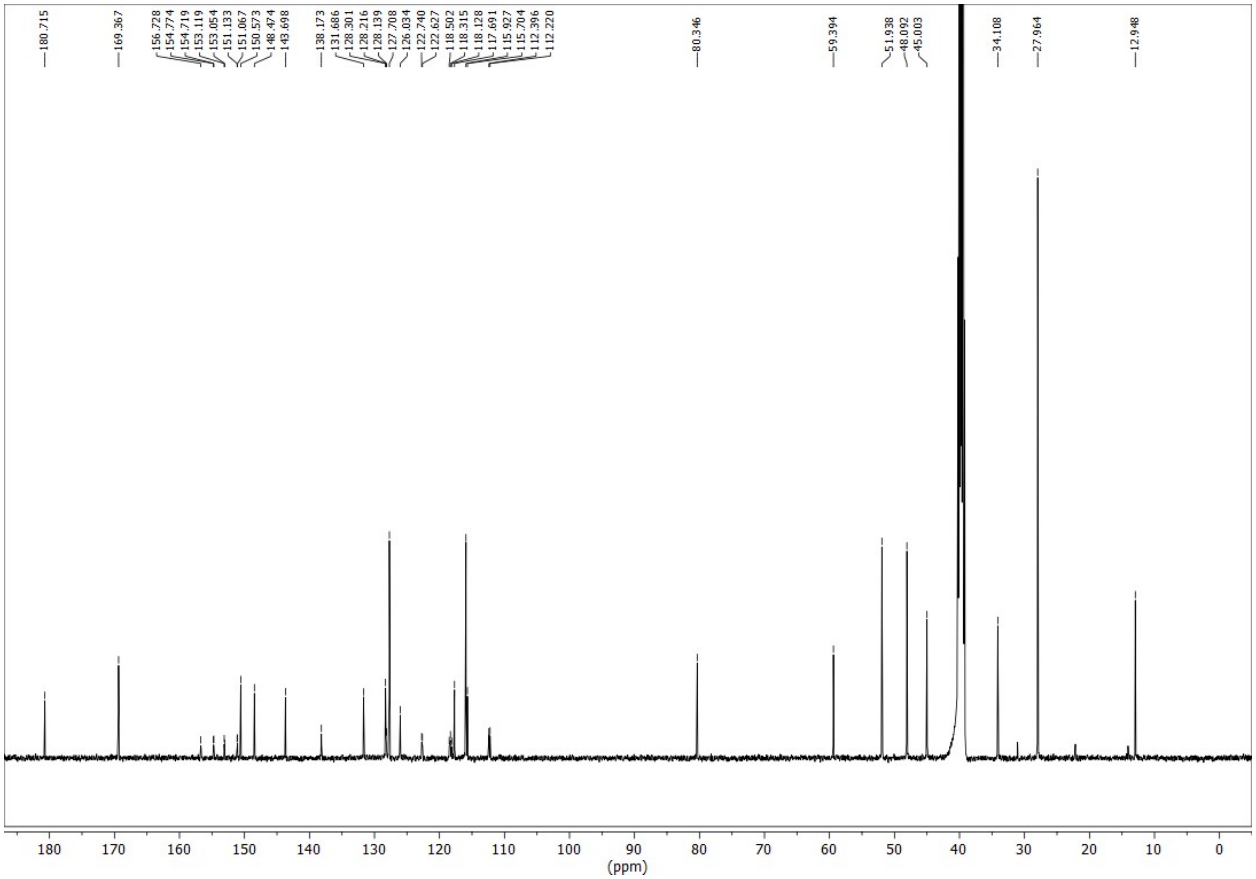
^{1}H and ^{13}C NMR spectrum of compound **21**.



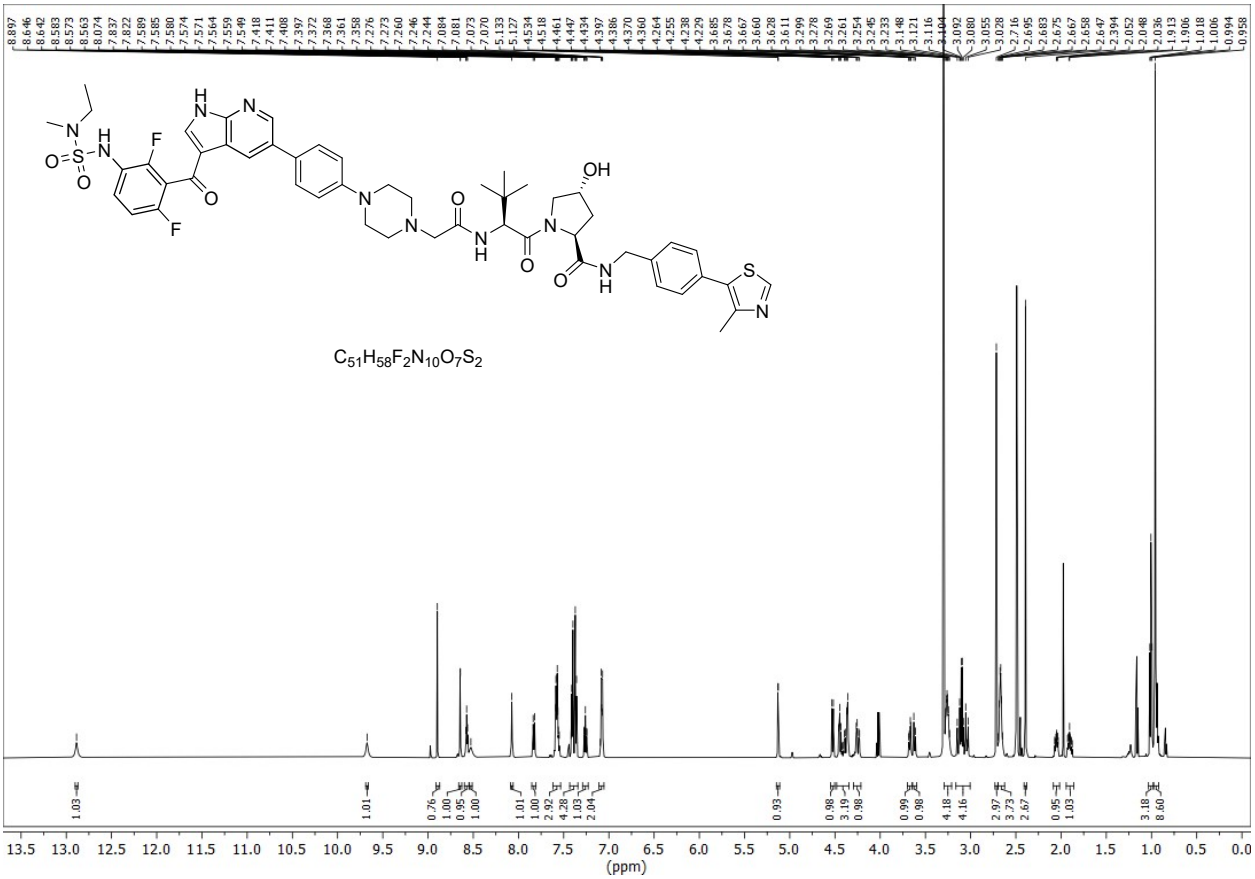


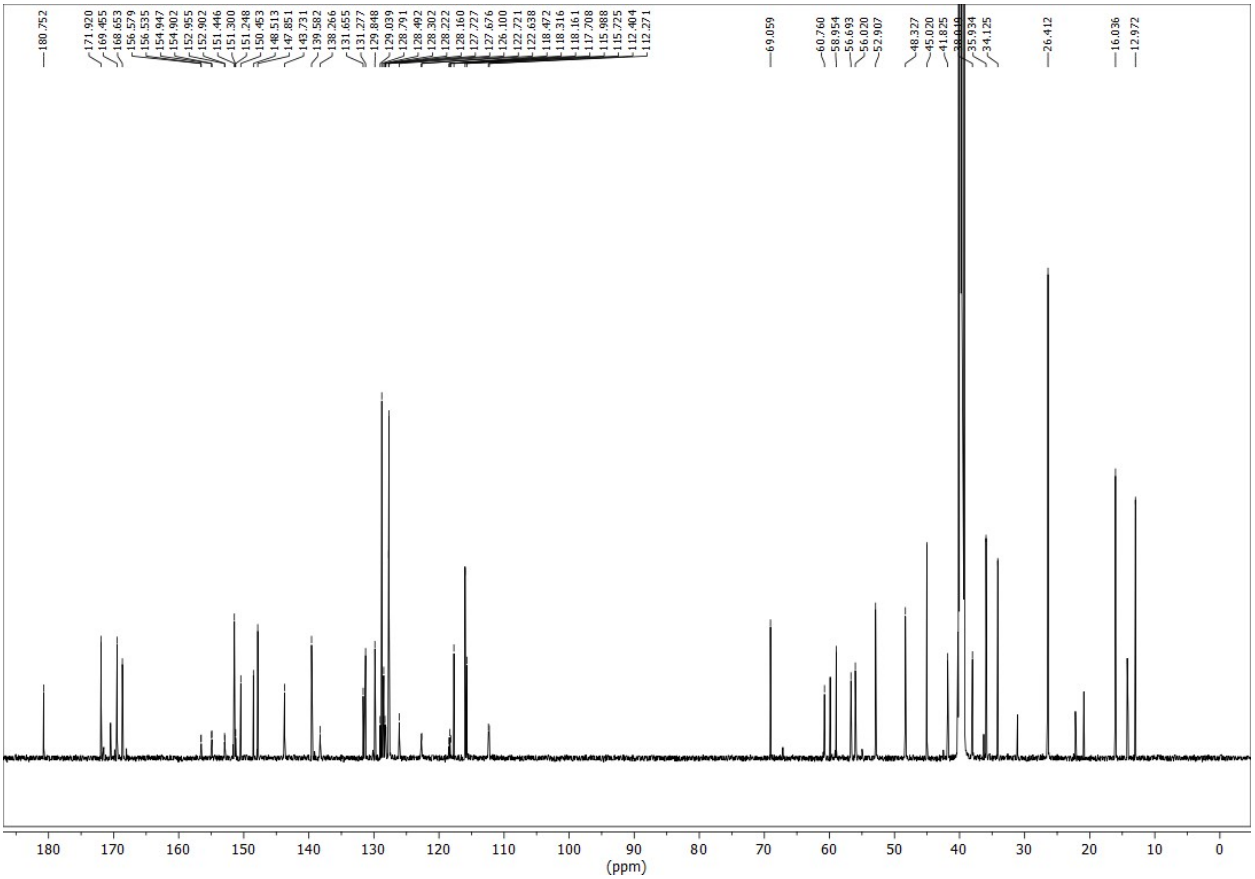
^{1}H and ^{13}C NMR spectrum of compound **23**.





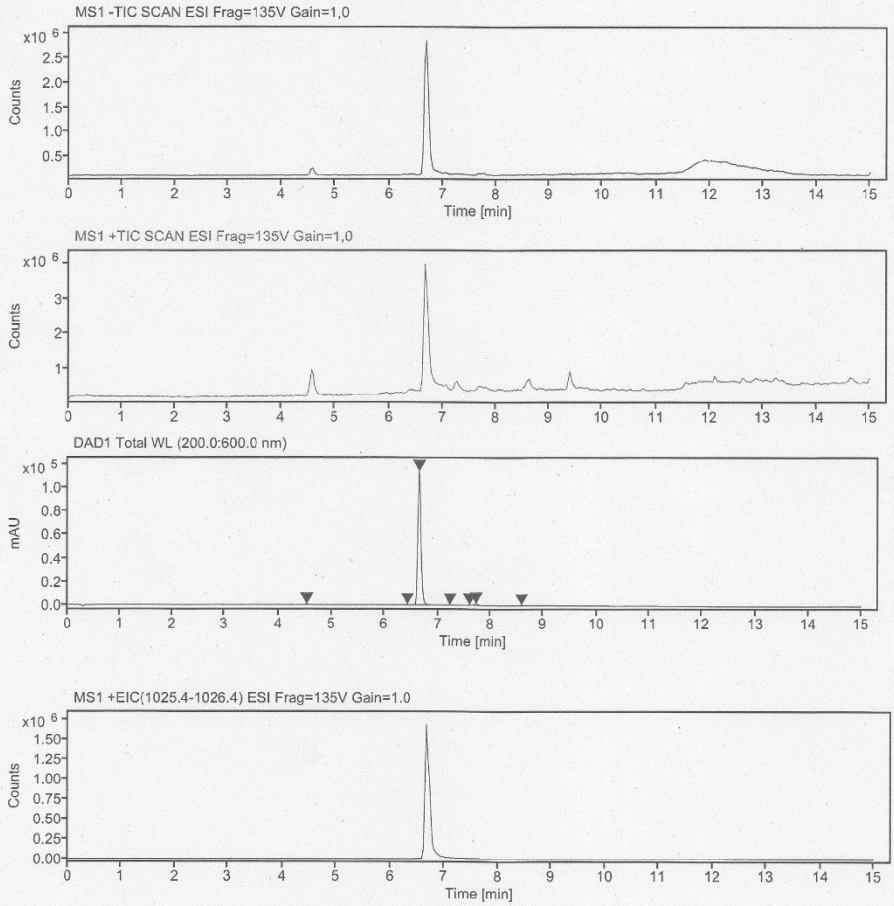
^{1}H and ^{13}C NMR spectrum of compound **25** (CST905).





LC/MS trace of compound **25** (CST905).

Sequence Name:	2021-05-20		
Data file:	CST905_ACN.dx	**Project Name:**	Single Quad
Sample name:	CST905_ACN	**Operator :**	SYSTEM
Instrument:	Single Quad	**Acquired on:**	2021-05-20 13:17:01+02:00
Inj. volume:	4.000	**Location:**	P2-C2
Acq. method:	0_wasser-acn_standard_100-1500_AC-20min.amx		
Processing method:	MS_standard_plot.pmx		



Signal: DAD1 Total WL (200.0:600.0 nm)

RT [min]	Peak MS Base Peak m/z	Area	Area%	Max Peak%	Height
4.533		2107.1410	0.4433	0.453	475.278
6.427		394.5716	0.0830	0.085	53.132
6.652		464872.5762	97.7948	100.000	114456.155
7.231		612.1168	0.1288	0.132	103.973
7.596		640.2079	0.1347	0.138	232.422
7.719		6375.9051	1.3413	1.372	1392.385
8.586		352.5678	0.0742	0.076	63.619
	Sum	475355.0864			

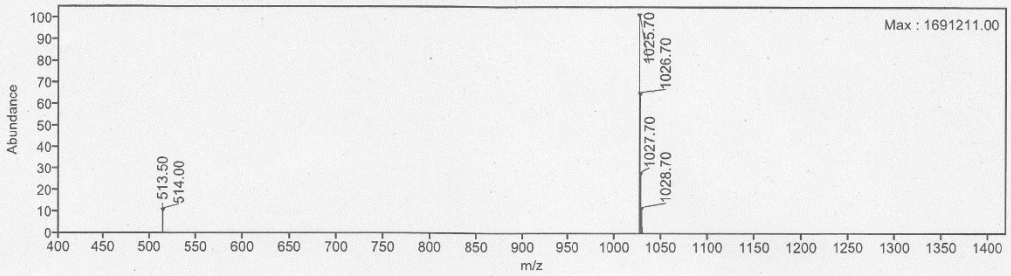
Signal: MS1 +TIC SCAN ESI Frag=135V Gain=1,0

RT [min]	Peak MS Base Peak m/z	Area	Area%	Max Peak%	Height
6.684	1025.700	17998135.1815	100.0000	100.000	3439289.954
	Sum	17998135.1815			

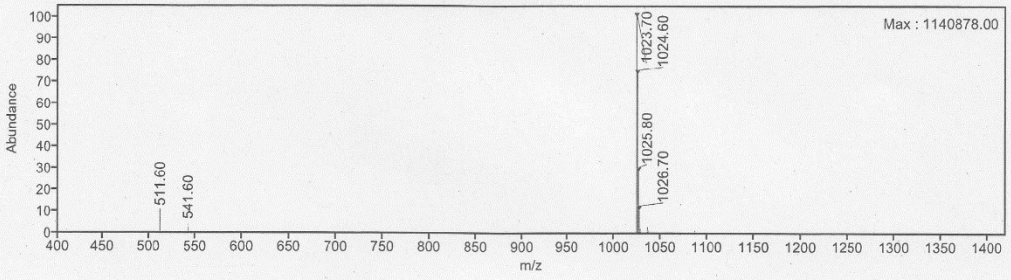
Signal: MS1 -TIC SCAN ESI Frag=135V Gain=1,0

RT [min]	Peak MS Base Peak m/z	Area	Area%	Max Peak%	Height
6.690	1023.700	14988115.2685	100.0000	100.000	2707454.474
	Sum	14988115.2685			

MS1 +TIC SCAN ESI Frag=135V Gain=1,0RT:6.68426025935167min



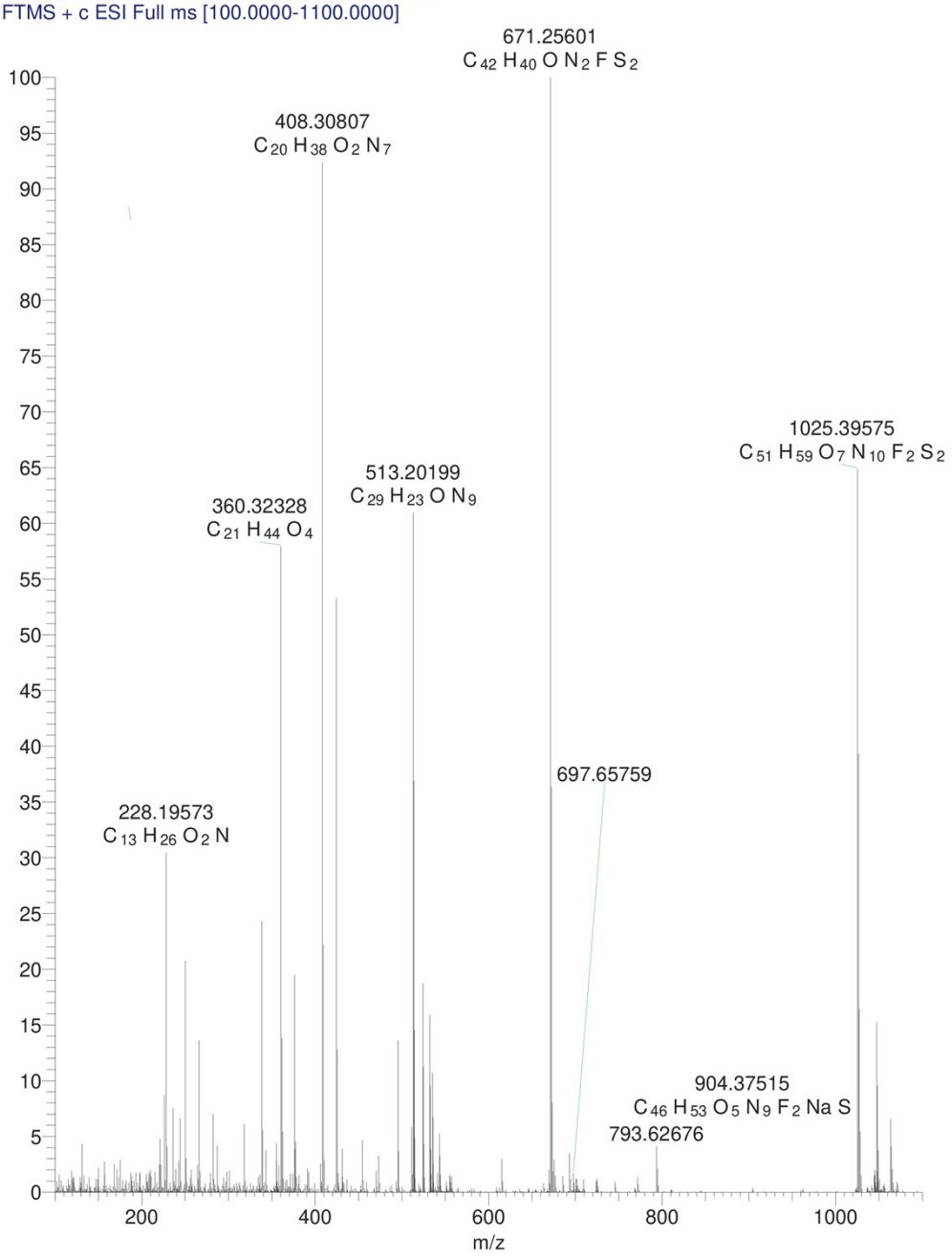
MS1 -TIC SCAN ESI Frag=135V Gain=1,0RT:6.69025695773485min



HRMS spectrum of compound **25** (CST905).

CST905

CST905 #21-29 RT: 0.09-0.13 AV: 9 NL: 1.40E7
T: FTMS + c ESI Full ms [100.0000-1100.0000]



Elemental composition search on mass 1025.39575

m/z= 1020.39575-1030.39575

m/z	Theo. Mass	Delta (ppm)	RDB equiv.	Composition
1025.39575	1025.39722	-1.43	26.5	C51 H59 O7 N10 F2 S2

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